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ORIGINAL ARTICLES

THE GENUS *FUSARIUM*

III. A CRITICAL STUDY OF THE FUNGUS CAUSING WILT OF GRAM (*CICER ARIETINUM* L.) AND OF THE RELATED SPECIES IN THE SUB-SECTION ORTHOCERA, WITH SPECIAL RELATION TO THE VARIABILITY OF KEY CHARACTERISTICS.

BY

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(Received for publication on 21 November 1939)

INTRODUCTION

IN the second contribution of this series, Prasad and Padwick [1939] described briefly the fungus causing gram wilt, and gave reasons for placing it in the sub-section Orthocera of the main section Elegans in the genus *Fusarium*. They pointed out that it was impossible to make any more definite statement without carefully comparing the fungus with all known species of the sub-section and without a thorough understanding of the range of variability of the isolates. The necessary study has now been made and the many facts established are held to throw considerable light not only on this particular fungus but also on the whole sub-section to which it belongs. The section Elegans is of great importance from plant pathological considerations. It has perhaps eight representatives so far known in India some, if not all of which cause havoc amongst cultivated crops. A thorough understanding of the range of variability of these fungi is necessary if we are to place identifications, and therefore breeding for disease resistance, on a sure footing. It is for this reason that the sub-section Orthocera has been subjected to such an elaborate study.

The sub-section Orthocera has, according to the classification of Wollenweber and Reinking [1935], five species, and with varieties and forms the total number of representatives is twelve. Eleven of these were secured for this work from the Centraalbureau voor Schimmelcultures, Baarn, Holland, the remaining representative being *Fusarium conglomerans* var. *citrinum* Wr. (= *Fusarium citrinum* Wr.) which was not obtainable at Baarn and which Wollenweber no longer has in his collection. These, together with the gram wilt fungi and a species isolated from a wilting linseed plant (*Linum usitatissimum* L.) from Karnal, Punjab, were the fungi studied. The complete list of fungi is therefore as follows:—

F. bostrycoides Wr. and Rkg. (Baarn)

F. conglomerans Wr. (Baarn)

F. conglomerans Wr. var. *betae* Stewart (Baarn)

F. conglomerans Wr. var. *callistephi* Beach (Baarn)

F. orthoceras App. and Wr. (Baarn)

F. orthoceras App. and Wr. var. *apii* (Nelson and Cochran) Wr. and Rkg. (= *F. apii* Nelson and Sherbakoff) (Baarn)

F. orthoceras App. and Wr. var. *apii* f. 1 Wr. and Rkg. (= *F. apii* var. *pallidum* Nelson and Sherbakoff) (Baarn)

F. orthoceras App. and Wr. var. *pisi* Linford (Baarn)

F. orthoceras App. and Wr. var. *longuis* (Sherb.) Wr. (Baarn)

F. angustum Sherb. (Baarn)

F. lini Bolley (Baarn)

F. lini Bolley? (isolated from Karnal, Punjab)

Fusarium species causing gram wilt (here designated F57, but called 'Type 2' by Prasad and Padwick)

Fusarium species causing gram wilt (here designated F92, but called 'Type 7' by Prasad and Padwick)

Fusarium species causing gram wilt (here designated F93, but called 'Type 8' by Prasad and Padwick)

The key to the identity of *Orthocera* *Fusaria* given by Wollenweber and Reinking omits *F. lini*. The characters necessary for distinguishing these fungi are as follows :—

- (1) Presence or absence of pionnotes.
- (2) Type of conidiophores (with bostrychoid branching or simple to branched in whorls).
- (3) Colour of stroma (pale, brownish white to flesh-coloured or red, violet, reddish brown or rust-red).
- (4) Type of plectenchyma—erumpent or smooth.
- (5) Sizes of conidia.
- (6) Pathogenicity.

These characters have been studied with the exception of the type of conidiophores (no sign of bostrychoid branching was found) and pathogenicity, a character which had to be taken for granted. Four experiments were conducted :

- (1) The effect of different plant extract media, commonly used in the identification of species of *Fusarium*, on the key characters.
- (2) The effect of temperature on the key characters.
- (3) A study of pigment production by *Fusarium orthoceras* var. *apii* f. 1.
- (4) The influence of asparagine on the key characters.

THE EFFECT OF DIFFERENT PLANT EXTRACT MEDIA, COMMONLY USED IN THE IDENTIFICATION OF SPECIES OF *FUSARIUM*, ON THE KEY CHARACTERS

The media used in this experiment were the following :—

Two per cent Potato Dextrose Agar.

Five per cent Potato Dextrose Agar.

Potato cylinders.

Steamed rice.

The media were prepared in the precise manner indicated by Wollenweber, Sherbakoff, Reinking, Johann and Bailey [1925], except that rice had to be steamed more than three times as proposed by these workers, in order to ensure complete sterilization under Indian conditions.

Cultures of the fungi were prepared on oatmeal agar for use in inoculating the agar slants of different media. When sufficiently grown, transfers were made of small portions of agar and mycelium to the four media, duplicate tubes being prepared.

It was intended to keep the tubes at a constant temperature of 20°C, but since at the time the difference between day and night temperature in the laboratory was sometimes as much as 18°C and only an ice-box was available for the purpose, great accuracy was not possible. During the first few days the temperature of the tubes was regularly 20°C in the morning but rose to 24° or 25°C for a brief period in the evening. After the first week the temperature remained steady at 19½° to 20½°C for the next twenty days, by which time observations were completed excepting a few notes of minor importance on colours of colonies and substrata.

Colours of the aerial mycelium and the substrate (i.e. the agar surface) were noted on the ninth and twenty-first days after inoculation. They are recorded in Tables I to IV. On the forty-fourth day the colours of the rice substrate were again noted. No change had occurred since the twenty-first day. Sufficient quantity of two per cent KOH solution was then passed into the tubes to cover the agar slope. The following day the colours were again noted. They are recorded also in Table I. The colour nomenclature of Ridgway [1912] was used throughout this work. Notes on abundance of aerial mycelium, presence or absence of a 'stroma', and type of conidia in the aerial mycelium, taken on the 22nd to 24th days after inoculation, are recorded in Tables V to VIII. Measurements of fifty microconidia of each culture on potato dextrose agar were made on the nineteenth and twentieth days. These measurements are given in Table IX.

The following are the conclusions of main importance drawn from experiment 1:—

- (1) Apart from (i) pale Russian blue aerial mycelium produced by *F. orthoceras* var. *pisi* on two per cent potato dextrose agar, and (ii) certain very pale and indefinite colours such as, ivory yellow, sea-shell pink, salmon buff, etc., all the cultures produced white aerial mycelium and a white or pale colour on the surface of the substrate or plectenchymatous stroma, or else produced a purple or closely related hue varying in tone and shade in both aerial mycelium and substrate, which always became plum purple or of a bluish violet hue on addition of KOH. The fungi thus fall in three groups:—
 - (a) The blue pigmented fungus. *F. orthoceras* var. *pisi*.
 - (b) The purplish pigmented group, including *F. bostrycoides*, *F. orthoceras*, *F. orthoceras* var. *apii*, *F. orthoceras* var. *longuis*, *F. angustum*, *F. lini*.
 - (c) The non-pigmented group, including *F. conglutinans*, *F. conglutinans* var. *betae*, *F. conglutinans* var. *callistephi*, *F. orthoceras* var. *apii* f.1, the gram wilt organisms.

Thus whereas all varieties of *F. conglutinans* were non-pigmented, the five forms of *F. orthoceras* fell within three groups.

- (2) The pigments were produced best on rice. On five per cent potato dextrose agar only *F. angustum* and *F. lini* produced a definite pigment, on two per cent potato dextrose agar only *F. lini* produced it, and potato cylinders were quite unsuitable for pigment production.

- (3) The colours present on the twenty-first day were generally speaking only an intensification of those on the ninth day.
- (4) The fungi varied greatly in amount of aerial mycelium produced. Potato cylinders and five per cent potato dextrose agar were generally speaking very unfavourable for its production, though three cultures produced abundant mycelium on the former and two cultures which produced none on the former produced abundant aerial mycelium on the latter. Within the five varieties or forms of *F. orthoceras* and the three varieties of *F. conglutinans* there was no relationship at all as regards abundance of aerial mycelium.
- (5) A true 'stroma' was absent throughout, but a plectenchymatous mat of hyphae reminiscent of a stroma sometimes occurred. This was not produced on rice mush or potato cylinders. On two per cent potato dextrose agar it was produced by the three varieties of *F. conglutinans* and the three gram wilt organisms, and on five per cent potato dextrose agar by the three varieties of *F. conglutinans*, the three gram wilt organisms, *F. orthoceras* var. *pisi* and *F. orthoceras* var. *longuis*.
- (6) Steamed rice was not suitable for producing conidia in the aerial mycelium. *F. orthoceras* var. *pisi* produced conidia in the aerial mycelium only on two per cent potato dextrose agar, and then only a few spores. All the other cultures behaved more or less alike, and all the media except rice were suitable for production of spores in the aerial mycelium whenever the latter was formed.
- (7) All the cultures were alike on all media in the following characteristics :—
 - (i) None produced chlamydospores by the twenty-second to twenty-fourth days after inoculation.
 - (ii) None produced sclerotia.
 - (iii) None produced sporodochia or pionnotes.
 - (iv) The only conidia produced were those in the aerial mycelium and they were continuous, ovoid to spindle-shaped or slightly curved, rounded equally at both ends, and borne singly (not in false heads or chains).

These facts have therefore not been recorded in the tables.

As regards measurements of spores, there was a highly significant difference between the longest spored isolate F 93 (the gram wilt fungus), and the shortest spored, F 85 (*F. orthoceras* var. *apii* f.1). The figures are as follows :—

Isolate	Mean length (μ)	S. E.	Difference	S. E. of difference
F 85 . . .	7.0	0.21	4.0 μ	0.67 μ
F 93 . . .	11.0	0.64		

If we take the means of the *authentic* cultures only, we have F 85 as the shortest spored isolate, and F 84 as the longest spored, the figures being as follows :—

Isolate	Mean length (μ)	S. E.	Difference	S. E. of difference
F 85 . . .	7.0	0.21	3.7 μ	0.51 μ
F 84 . . .	10.7	0.46		

This difference is again highly significant, and it will be noted that both F 84 and F 85 are forms of *F. orthoceras* var. *apii*. It may be added that the gram wilt organism F 93 had significantly larger conidia than the other two gram wilt organisms, F 57 and F 92.

TABLE I

Colours of aerial mycelium and surface of substrate of rice cultures on the ninth and twenty-first days, and after adding potassium hydroxide on the forty-fourth day

Culture	Aerial mycelium		Surface of Substrate		
	9th day	21st day	9th day	21st day	44th day (with KOH)
F 79 <i>F. bestrycoides</i>	White . .	White . .	Vinaceous drab and Payne's gray	Vinaceous drab .	Blue-violet black
F 80 <i>F. conglutinans</i>	White . .	White . .	Seashell pink .	White and seashell pink	White
F 81 <i>F. conglutinans</i> var. <i>betae</i>	White . .	White . .	Unchanged .	Unchanged .	Honey yellow
F 82 <i>F. conglutinans</i> var. <i>callistephi</i>	White . .	White . .	Do. .	Ivory yellow .	Isabella colour
F 83 <i>F. orthoceras</i>	White . .	White and old rose	Orange vinaceous	Perilla purple .	Dull bluish violet
F 84 <i>F. orthoceras</i> var. <i>apii</i>	White . .	White and old rose	Light perilla purple and light pinkish lilac	Dark perilla purple and light perilla purple	Dark dull bluish violet
F 85 <i>F. orthoceras</i> var. <i>apii</i> f.1	White . .	White . .	Salmon buff .	Salmon buff .	White
F 73 <i>F. orthoceras</i> var. <i>pisi</i>	White . .	White . .	Unchanged .	Unchanged .	Deep mouse grey
F 86 <i>F. orthoceras</i> var. <i>longius</i>	White . .	White, hellebore red and argyle purple	Light perilla purple and light pinkish lilac	Dark perilla purple to light pinkish lilac	Plum purple
F 87 <i>F. angustum</i>	White . .	White, hellebore red and argyle purple	Light perilla purple and rhodonite pink	Dark perilla purple to bishop's purple	Plum purple
F 74 <i>F. lini</i>	White and purplish lilac	White and purplish lilac	Light perilla purple	Purplish lilac .	Plum purple
F 20 <i>F. lini</i> ? (Karnal)	White . .	White and orange vinaceous	Perilla purple and light pinkish lilac	Dark perilla purple	Plum purple
F 57 Gram wilt organism	White . .	White . .	Unchanged .	Unchanged .	Seafoam green
F 92 Gram wilt organism	White . .	White . .	Unchanged .	Unchanged .	Cream buff
F 93 Gram wilt organism	White . .	White . .	Unchanged .	Unchanged .	White

TABLE II

Colour of aerial mycelium and surface of substrate of potato cylinder cultures on the ninth and twenty-first days

Culture	Aerial mycelium		Surface of substrate	
	9th day	21st day	9th day	21st day
F 79 <i>F. bostrycoides</i>	White	White	Pale olive buff	Ivory yellow
F 80 <i>F. conglutinans</i>	White	White	Ivory yellow	Ivory yellow
F 81 <i>F. conglutinans</i> var. <i>betae</i>	White	White	Ivory yellow	Ivory yellow
F 82 <i>F. conglutinans</i> var. <i>calistephi</i>	White	White	Ivory yellow	Ivory yellow
F 83 <i>F. orthoceras</i>	Lacking	Lacking	Ivory yellow	Ivory yellow
F 84 <i>F. orthoceras</i> var. <i>apii</i>	White and light pinkish lilac	Lacking	Ivory yellow	Ivory yellow
F 85 <i>F. orthoceras</i> var. <i>apii</i> f. 1	Lacking	Lacking	Ivory yellow	Ivory yellow
F 73 <i>F. orthoceras</i> var. <i>pisi</i>	White	White	White	Colourless
F 86 <i>F. orthoceras</i> var. <i>longuis</i>	White and light pinkish lilac	Lacking	Ivory yellow	Ivory yellow
F 87 <i>F. angustum</i>	White and ageratum violet	White and ageratum violet	Ivory yellow	Ivory yellow
F 74 <i>F. lini</i>	White with traces of purplish black	White	Ivory yellow	Ivory yellow and argyle purple
F 20 <i>F. lini</i> ? (Karnal)	White and purplish lilac	White and purplish lilac	Ivory yellow	Ivory yellow
F 57 Gram wilt organism	White	White	Unchnaged	Ivory yellow
F 92 Gram wilt organism	White	White	Unchanged	Unchanged
F 93 Gram wilt organism	White	White	Ivory yellow	Ivory yellow

TABLE III

Colour of aerial mycelium and surface of substrate of two per cent potato dextrose agar cultures on the ninth and twenty-first days

Culture	Aerial mycelium		Surface of substrate	
	9th day	21st day	9th day	21st day
F 79 <i>F. bostrycoides</i>	White	White	Pale olive buff	Ivory yellow
F 80 <i>F. conglutinans</i>	White	White and glaucous blue	White	White and pale glaucous blue
F 81 <i>F. conglutinans</i> var. <i>betae</i>	Pale olive buff	White	Pale olive buff	Pale olive buff
F 82 <i>F. conglutinans</i> var. <i>calistephi</i>	White	White	Ivory yellow	Ivory yellow
F 83 <i>F. orthoceras</i>	White	White	White	Ivory yellow
F 84 <i>F. orthoceras</i> var. <i>apii</i>	White	White and pale salmon colour	Ivory yellow	Ivory yellow
F 85 <i>F. orthoceras</i> var. <i>apii</i> f. 1	White	White	White	White
F 73 <i>F. orthoceras</i> var. <i>pisi</i>	White and light gull grey	White and pale Russian blue	White	Deep bluish gray green
F 86 <i>F. orthoceras</i> var. <i>longuis</i>	White	White and shell pink.	White	White
F 87 <i>F. angustum</i>	White and light pinkish lilac	White and purplish lilac	White	White
F 74 <i>F. lini</i>	White and purplish lilac	White and purplish lilac	Purplish lilac	Ivory yellow and purplish lilac
F 20 <i>F. lini</i> ? (Karnal)	White and light perilla purple	White and light perilla purple	White	Ivory yellow
F 57 Gram wilt organism	White and salmon buff	White	Ivory yellow	Ivory yellow
F 92 Gram wilt organism	White	White	White	Unchanged
F 93 Gram wilt organism	White	White	White	Unchanged

TABLE IV

Colour of aerial mycelium and surface of substrate of five per cent potato dextrose agar cultures on the ninth and twenty-first days

Culture	Aerial mycelium		Surface of substrate	
	9th day	21st day	9th day	21st day
F 79 <i>F. bostrycoides</i> . . .	White . . .	White . . .	Pale olive buff . .	White
F 80 <i>F. conglutinans</i> . . .	White . . .	White and pale glaucous blue	White . . .	White and pale glaucous blue
F 81 <i>F. conglutinans</i> var. <i>betae</i>	Pale olive buff . .	White . . .	Pale olive buff . .	Pale olive buff
F 82 <i>F. conglutinans</i> var. <i>callistephi</i>	White . . .	White . . .	Ivory yellow . . .	Ivory yellow
F 83 <i>F. orthoceras</i> . . .	White . . .	White . . .	White . . .	Ivory yellow
F 84 <i>F. orthoceras</i> var. <i>apii</i> . .	White . . .	White and pale salmon colour	Ivory yellow . .	Ivory yellow
F 85 <i>F. orthoceras</i> var. <i>apii</i> f.1	White . . .	White . . .	White . . .	White
F 73 <i>F. orthoceras</i> var. <i>pisi</i> . .	White . . .	White . . .	White . . .	White
F 86 <i>F. orthoceras</i> var. <i>longuis</i>	White . . .	White . . .	White . . .	White
F 87 <i>F. angustum</i> . . .	White and light pinkish lilac	White and purplish lilac	White . . .	White
F 74 <i>F. lini</i> . . .	White and purplish lilac	White and purplish lilac	Purplish lilac . .	Ivory yellow and purplish lilac
F 20 <i>F. lini</i> ? (Karnal) . . .	White, light perilla purple and vinaceous pink	White, light perilla purple and vinaceous pink	White . . .	Ivory yellow
F 57 Gram wilt organism . . .	White . . .	Lacking . . .	Ivory yellow . .	Ivory yellow
F 92 Gram wilt organism . . .	White . . .	White . . .	White . . .	Unchanged
F 93 Gram wilt organism . . .	White . . .	White . . .	White . . .	White

TABLE V

Aerial mycelium, 'stroma' and conidia in aerial mycelium, on rice on the twenty-second to twenty-fourth days

Culture	Aerial mycelium	'Stroma'	Conidia in aerial mycelium
F 79 <i>F. bostrycoides</i> . . .	Moderate . .	Absent . .	Vacuolated, very abundant
F 80 <i>F. conglutinans</i> . . .	Moderate . .	Absent . .	Hyaline, few
F 81 <i>F. conglutinans</i> var. <i>betae</i>	Moderate . .	Absent . .	Absent
F 82 <i>F. conglutinans</i> var. <i>callistephi</i>	Scanty . . .	Absent . .	Vacuolated, very abundant
F 83 <i>F. orthoceras</i> . . .	Scanty . . .	Absent . .	Vacuolated, few
F 84 <i>F. orthoceras</i> var. <i>apii</i> . .	Moderate . .	Absent . .	Vacuolated, moderately abundant
F 85 <i>F. orthoceras</i> var. <i>apii</i> f.1	Scanty . . .	Absent . .	Absent
F 73 <i>F. orthoceras</i> var. <i>pisi</i> . .	Moderate . .	Absent . .	Absent
F 86 <i>F. orthoceras</i> var. <i>longuis</i>	Moderate . .	Absent . .	Highly vacuolated, few
F 87 <i>F. angustum</i> . . .	Moderate . .	Absent . .	Highly vacuolated, rare
F 74 <i>F. lini</i> . . .	Moderate . .	Absent . .	Highly vacuolated, moderately abundant
F 20 <i>F. lini</i> ? (Karnal) . . .	Moderate . .	Absent . .	Vacuolated, abundant
F 57 Gram wilt organism . . .	Scanty . . .	Absent . .	Sometimes constricted at middle, vacuolated, moderately abundant
F 92 Gram wilt organism . . .	Moderate . .	Absent . .	Hyaline, few
F 93 Gram wilt organism . . .	Scanty . . .	Absent . .	Various distorted shapes, with swellings and sharp bends, hyaline, moderately abundant.

TABLE VI

Aerial mycelium, 'stroma', and conidia in aerial mycelium, on potato cylinders on the twenty-second to twenty-fourth days

Culture	Aerial mycelium	'Stroma'	Conidia in aerial mycelium
F 79 <i>F. bostrycoides</i> . . .	Scanty .	Absent .	Vacuolated, very abundant
F 80 <i>F. conglomerans</i> . . .	Moderate .	Absent .	Highly vacuolated, abundant
F 81 <i>F. conglomerans</i> var. <i>betae</i>	Abundant	Absent .	Hyaline, moderately abundant
F 82 <i>F. conglomerans</i> var. <i>callistephi</i> .	Absent .	Absent .	Vacuolated, very abundant
F 83 <i>F. orthoceras</i> . . .	Absent .	Absent .	Highly vacuolated, abundant
F 84 <i>F. orthoceras</i> var. <i>apii</i> .	Absent .	Absent .	Highly vacuolated, very abundant
F 85 <i>F. orthoceras</i> var. <i>apii</i> f.1	Absent .	Absent .	Hyaline, moderately abundant
F 73 <i>F. orthoceras</i> var. <i>pisi</i> .	Moderate .	Absent .	Absent
F 86 <i>F. orthoceras</i> var. <i>longius</i>	Absent .	Absent .	Highly vacuolated, very abundant
F 87 <i>F. angustum</i> . . .	Absent .	Absent .	Highly vacuolated, moderately abundant
F 74 <i>F. lini</i> . . .	Abundant	Absent .	Highly vacuolated, moderately abundant
F 20 <i>F. lini</i> ? (Karnal). .	Absent .	Absent .	Sometimes constricted at middle, highly vacuolated, abundant
F 57 Gram wilt organism .	Absent .	Absent .	Sometimes constricted at middle, vacuolated, very abundant
F 92 Gram wilt organism .	Abundant	Absent .	Hyaline, moderately abundant
F 93 Gram wilt organism .	Absent .	Absent .	Highly vacuolated, moderately abundant

TABLE VII

Aerial mycelium, 'stroma', and conidia in aerial mycelium, on two per cent potato dextrose agar on the twenty-second to twenty-fourth days

Culture	Aerial mycelium	'Stroma'	Conidia in aerial mycelium
F 79 <i>F. bostrycoides</i> .	Moderate .	Absent . .	Vacuolated, very abundant
F 80 <i>F. conglutinans</i> .	Scanty . .	Plectenchymatous	Hyaline, few
F 81 <i>F. conglutinans</i> var. <i>betae</i>	Scanty . .	Plectenchymatous	Vacuolated, few
F 82 <i>F. conglutinans</i> var. <i>callistephi</i>	Scanty . .	Plectenchymatous	Vacuolated, very abundant
F 83 <i>F. orthoceras</i> .	Moderate .	Absent . .	Hyaline, abundant
F 84 <i>F. orthoceras</i> var. <i>apii</i> .	Abundant .	Absent . .	Somewhat vacuolated, very abundant
F 85 <i>F. orthoceras</i> var. <i>apii</i> f. 1	Moderate .	Absent . .	Sometimes constricted at middle, vacuolated moderately abundant
F 73 <i>F. orthoceras</i> var. <i>pisi</i>	Moderate .	Absent . .	Hyaline, few
F 86 <i>F. orthoceras</i> var. <i>longuis</i>	Abundant .	Absent . .	Hyaline, very abundant
87 <i>F. angustum</i> .	Moderate .	Absent . .	Hyaline, moderately abundant
F 74 <i>F. lini</i> . .	Moderate .	Absent . .	Highly vacuolated, few
F 20 <i>F. lini</i> ? (Karnal)	Moderate .	Absent . .	Hyaline, abundant
F 57 Gram wilt organ- ism	Scanty . .	Plectenchymatous	Hyaline, very abundant
F 92 Gram wilt organ- ism	Moderate .	Plectenchymatous	Hyaline, moderately abundant
F 93 Gram wilt organ- ism	Short and thick	Plectenchymatous	Somewhat vacuolated, moderately abundant

TABLE VIII

Aerial mycelium, 'stroma,' and conidia in aerial mycelium, on five per cent potato dextrose agar on the twenty-second to twenty-fourth days

Culture	Aerial mycelium	'Stroma'	Conidia in aerial mycelium
F 79 <i>F. bostrycoides</i> .	Scanty . .	Absent . .	Vacuolated, very abundant
F 80 <i>F. conglutinans</i> .	Scanty . .	Plectenchymatous	Hyaline, few
F 81 <i>F. conglutinans</i> var. <i>betae</i>	Scanty . .	Plectenchymatous	Vacuolated, few
F 82 <i>F. conglutinans</i> var. <i>callistephi</i>	Absent . .	Plectenchymatous	Spores often constricted at middle and distorted, vacuolated, abundant
F 83 <i>F. orthoceras</i> .	Scanty . .	Absent . .	Spores often constricted at middle and distorted, vacuolated, abundant
F 84 <i>F. orthoceras</i> var. <i>apii</i>	Abundant .	Absent . .	Hyaline, very abundant
F 85 <i>F. orthoceras</i> var. <i>apii</i> f.1	Scanty . .	Absent . .	Vacuolated, few
F 73 <i>F. orthoceras</i> var. <i>pisi</i>	Scanty . .	Plectenchymatous	Absent
F 86 <i>F. orthoceras</i> var. longuis	Abundant .	Plectenchymatous	Hyaline very abundant
F 87 <i>F. angustum</i> .	Scanty . .	Absent . .	Hyaline, moderately abundant
F 74 <i>F. lini</i> . .	Moderate .	Absent . .	Absent
F 20 <i>F. lini</i> ? (Karnal)	Moderate .	Absent . .	Slightly vacuolated, abundant
F 57 Gram wilt organism	Scanty . .	Plectenchymatous	Hyaline, moderately abundant
F 92 Gram wilt organism	Scanty . .	Plectenchymatous	Hyaline, moderately abundant
F 93 Gram wilt organism	Scanty . .	Plectenchymatous	Hyaline, few

TABLE IX

Measurements of microconidia on two per cent potato dextrose agar on the nineteenth and twentieth days (means of fifty conidia)

Culture	Mean length (μ)	Mean breadth (μ)	Range (μ)
F 79 <i>F. bostrycoides</i> . .	8.6	3.5	5.1-14.3 × 1.7-5.1
F 80 <i>F. conglutinans</i> . .	9.9	3.4	6.8-18.0 × 2.0-4.4
F 81 <i>F. conglutinans</i> var. <i>betae</i>	Absent
F 82 <i>F. conglutinans</i> var. <i>callistephi</i>	10.3	2.4	7.8-19.4 × 1.7-4.1
F 83 <i>F. orthoceras</i> . .	9.0	3.2	5.4-15.0 × 2.4-4.4
F 84 <i>F. orthoceras</i> var. <i>apii</i> .	10.7	3.2	5.4-18.7 × 2.0-4.4
F 85 <i>F. orthoceras</i> var. <i>apii</i> f.1	7.0	3.0	4.4-10.2 × 1.7-4.1
F 73 <i>F. orthoceras</i> var. <i>pisi</i> .	Absent
F 86 <i>F. orthoceras</i> var. <i>longius</i>	9.4	2.8	4.4-19.7 × 1.7-4.1
F 87 <i>F. angustum</i> . .	8.3	3.1	5.1-15.3 × 1.7-4.4
F 74 <i>F. lini</i>	10.3	3.5	6.8-15.3 × 2.0-4.1
F 20 <i>lini</i> ? (Karnal) . .	9.1	2.9	6.1-15.3 × 1.7-4.1
F 57 Gram wilt organism .	8.2	3.5	4.1-15.3 × 1.7-5.1
F 92 Gram wilt organism .	8.2	3.2	3.4-16.3 × 2.0-6.8
F 93 Gram wilt organism .	11.0	3.4	5.1-26.5 × 2.0-7.8

EFFECT OF TEMPERATURE ON THE KEY CHARACTERS

The medium used in this experiment was oatmeal agar. It was prepared with only 30 grams of oatmeal to a litre of water, but in other respects it was prepared in a manner similar to that of Wollenweber, Sherbakoff, Reinking, Johann and Bailey. Twelve agar-slants were prepared from each isolate, six day old oatmeal cultures being used for the purpose. The cultures were divided into six lots, each containing two tubes of each isolate. The six lots were held at six different temperatures in dark incubators. After ten days notes were made of the colours of the surface of the substrate and the aerial mycelium, together with the amount of aerial mycelium. After a further ten days the colours were again noted. On the twentieth day note-taking began as regards other characters as well as colour production,

and it took six days to complete the work. These notes included the presence or absence of a stroma, the type of conidia on the surface of the medium, the presence or absence of a pionnotal layer, and the development of chlamydospores. It was not possible to take the more detailed observations at all temperatures, however, and consequently the temperatures 20° and 35°C were chosen for observations on conidia and chlamydospores except in one or two special cases where all temperatures were used.

The temperatures were recorded twice daily and the averages and ranges of the temperatures recorded during the period of the experiment were as follows :—

9.6°C (Range 8.5-11.0)

14.7°C (Range 14.0-16.0)

20.2°C (Range 17.5-23.0)

25.4°C (Range 25.0-26.5)

30.2°C (Range 27.5-32.0)

35.3°C (Range 34.5-36.5)

It will be noticed that the most difficult temperatures to maintain constant were those at 20, 25 and 30°C, which were nearest to room temperatures during the period. The means, however, were close to the required temperatures of 10, 15, 20, 25, 30 and 35°C, and throughout this paper the cultures growing in the various incubators are referred to as the 10° Series, 15° Series, etc. Since the purpose is to compare the reactions of the various cultures to all six different temperatures it has been necessary to arrange somewhat elaborate tables. The colour observations are recorded in Table X, and this table also has been utilized for the relative thickness of the plectenchymatous stromata. No sporodochia or sclerotia were found, but in the case of F 82 (*F. conglutinans* var. *callistephi*) there was a definite pionnotal layer on the surface of the agar at all temperatures, though it was very thin in the 10°C. In several other cases thin layers of spores were found on the agar surface, though they scarcely deserved the name pionnotes. These observations are also included in Table X.

Owing to the fact that the aerial mycelium had frequently collapsed by the twentieth day, observations on conidia were made on scrapings taken from the surface of the agar in many cases. If pionnotes or a thin layer of spores were present, these spores were also described. The descriptions of conidia and chlamydospores are given in Table XI. These descriptions are given only for the temperature series 20°C and 35°C. In many, though not all, cases, observations were made in some or all of the other temperature series, but since the descriptions given in Table XI clearly illustrate the general trend of the results and the other observations merely supplement the general conclusions, these observations have been omitted in order to simplify comparisons.

TABLE X

Colour of aerial mycelium and surface of substrate on the tenth and twentieth days, and thickness of plectenchymatous stroma and pionotal layers on the twentieth to twenty-third days, at different temperatures

Culture	Temperature series, °C	Aerial mycelium		Surface of substrate		'Stroma'	Pionotal layer
		10th day	20th day	10th day	20th day		
F 79 <i>F. bostryoides</i>	10	— (0)	White (1)	Unchanged	Unchanged	Absent	Absent
	15	White (1)	White (2)	Do.	Do.	Do.	Do.
	20	White (1)	White (2)	Do.	Do.	Do.	Do.
	25	White (2)	White (2)	Do.	Do.	Do.	Do.
	30	White (2)	White (2)	Do.	Do.	Do.	Do.
	35	White (2)	White (2)	Do.	Do.	Do.	Do.
F 80 <i>F. conglutinans</i>	10	— (0)	White (1)	Unchanged	Unchanged	Absent	Absent
	15	White (1)	White (2)	Do.	Do.	Thin	Do.
	20	White (1)	White (2)	Do.	Do.	Do.	Do.
	25	White (3)	White (2)	Do.	Do.	Do.	Do.
	30	White (2)	White (3)	Do.	Do.	Do.	Do.
	35	White (2)	White (2)	Do.	Do.	Do.	Do.
F 81 <i>F. conglutinans</i> var. <i>betae</i>	10	— (0)	White (1)	Unchanged	Unchanged	Absent	Absent
	15	— (0)	— (0)	Do.	Do.	Thin	Do.
	20	— (0)	White (1)	Do.	Do.	Do.	Do.
	25	White (1)	White (1)	Do.	Do.	Do.	Do.
	30	White (1)	White (1)	Do.	Do.	Do.	Do.
	35	White (1)	White (1)	Do.	Do.	Do.	Do.

TABLE X—*contd.*

Culture	Temperature series, °C	Aerial mycelium		Surface of substrate		'Stroma'	Pionotal layer
		10th day	20th day	10th day	20th day		
F 82 <i>F. conglutinans</i> var. <i>calisteph.</i>	10	— (0)	White (1)	Unchanged	Unchanged	Absent	? A very thin layer.
	15	— (0)	White (1)	Do.	Do.	Thin	Pionotes present
	20	— (0)	White (1)	Do.	Do.	Do.	Do.
	25	White (2)	White (1)	Do.	Cream buff	Do.	Do.
	30	White (1)	— (0)	Do.	Unchanged	Do.	Do.
	35	White (1)	— (0)	Do.	Do.	Do.	Do.
F 83 <i>F. orthoceras</i>	10	— (0)	Trace of purplish lilac (2)	Light perilla purple.	Light perilla purple.	Absent	Absent
	15	White (2)	Trace of purplish lilac (3)	Argyle purple	Light perilla purple.	Thin	Do.
	20	Trace of purplish lilac (3)	Trace of purplish lilac (3)	Bishop's purple	Perilla purple	Do.	Do.
	25	Trace of light purplish lilac (3).	Trace of Bishop's purple (3)	Light perilla purple.	Dark perilla purple	Do.	Do.
	30	Trace of purplish lilac (3)	Trace of purplish lilac (2)	Perilla purple	Perilla purple	Do.	Do.
	35	Trace of light lobelia violet (3).	Trace of purplish lilac (2)	Naphthalene violet.	Dark naphthalene violet.	Absent	Do.
F 84 <i>F. orthoceras</i> var. <i>apii.</i>	10	— (0)	Trace of purplish lilac (1)	Traces of light pinkish lilac.	Pale vinaceous lilac.	Absent	Absent
	15	White (1)	Trace of purplish lilac (2)	Purplish lilac	Light perilla purple.	Thin	? A very thin layer.
	20	Trace of purplish lilac (1)	Trace of purplish lilac (2)	Bishop's purple	Dark perilla purple	Do.	Do.
	25	Trace of light purplish lilac (3)	Trace of bishop's purple (3)	Light perilla purple.	Do.	Do.	Do.
	30	Trace of purplish lilac (3)	Trace of purplish lilac (2)	Perilla purple	Perilla purple	Do.	Absent
	35	Trace of light lobelia violet (3)	Trace of purplish lilac (2)	Naphthalene violet.	Dark naphthalene violet.	Do.	Do.

F 85 <i>F. orthoceras</i> var. <i>apii</i> , f. 1.	10	—	(0)	White (1)	Unchanged	Unchanged	Absent	Absent
	15	—	(0)	White (2)	Do.	Do.	Thin	Do.
	20	Trace of purplish lilac (2).		Bishop's purple (2)	Do.	Dark plumbeous	Do.	Do.
	25	Trace of lilac.*		Trace of purplish lilac (2).	Do.	Plumbeous and dark perilla purple.	Do.	Do.
	30	White (2).		White (1)	Do.	Unchanged	Do.	Do.
	35	White (3).		White (2)	Do.	Do.	Do.	Do.
F 73 <i>F. orthoceras</i> var. <i>pict.</i>	10	—	(0)	White (1)	Unchanged	Unchanged	Absent	Absent
	15	White (1).		White (2)	Do.	Trace of lilac buff.	Thin	Do.
	20	White (1).		White (2)	Do.	Onion-skin pink	Do.	Do.
	25	White (3).		White (2)	Do.	Do.	Do.	Do.
	30	White (3).		White (3)	Vinaceous cinnamon	Light russet vinaceous.	Do.	Do.
	35	White (3).		White (2)	Wood brown.	Dark vinaceous drab.	Do.	Do.
F 84 <i>F. orthoceras</i> var. <i>longist.</i>	10	—	(0)	Trace of purplish lilac (1).	Trace of light pinkish lilac	Pale vinaceous lilac	Absent	? A very thin layer
	15	Trace of purplish lilac (1).		Trace of purplish lilac (2).	Purplish lilac	Light perilla purple.	Thin	Absent
	20	Trace of purplish lilac (2).		Trace of purplish lilac (2).	Bishops' purple	Light perill purple.	Do.	A very thin layer
	25	Trace of light purplish lilac (3).		Trace of bishop's purple (3).	Light perilla purple.	Dark perilla purple.	Do.	Do.
	30	Trace of purplish lilac (3).		Trace of purplish lilac (2).	Perilla purple	Perilla purple	Do.	Do.
	35	Trace of light lobelia violet (3)		Trace of purplish lilac (2).	Nepthalene violet.	Dark naphthalene violet.	Do.	Do.
F 87 <i>F. angustum</i>	10	—	(0)	Trace of purplish lilac (2).	Purplish lilac	Light perilla purple.	Thin	Absent
	15	Trace of purplish lilac (2).		Trace of purplish lilac (3).	Argyle purple	Do.	Do.	Do.
	20	Trace of purplish lilac (3).		Trace of purplish lilac (3).	Light perilla purple.	Perilla purple	Do.	Do.
	25	Trace of light purplish lilac (3).		Trace of purplish lilac (3).	Do.	Do.	Do.	Do.
	30	Trace of purplish lilac (3).		Trace of purplish lilac (2).	Perilla purple	Do.	Do.	? A very thin layer.
	35	Trace of light lobelia violet (3)		Trace of purplish lilac (2).	Naphthalene violet.	Dark naphthalene violet.	Do.	Absent

TABLE X—*contd.*

Culture	Temperature series, °C	Aerial mycelium		Surface of substrate		'Stroma'	Punctal layer
		10th day	20th day	10th day	20th day		
F 74 <i>F. lini</i> . . .	10	— (0)	White (1)	Unchanged	Light pinkish lilac	Absent	Absent
	15	— (0)	Trace of purplish lilac (2)	Purplish lilac	Light perilla purple	Thin	Do.
	20	White (1)	Trace of purplish lilac (2)	Light perilla purple	Dark perilla purple	Do.	? A very thin layer
	25	Trace of light purplish lilac (3)	Trace of purplish lilac (2)	Do.	Do.	Do.	Absent
	30	White (2)	Trace of purplish lilac (2)	Perilla purple	Perilla purple	Do.	? A very thin layer
	35	White (3)	Trace of purplish lilac (3)	Dark hyssop violet	Dark naphthalene violet	Do.	Absent
F 20 <i>F. lini</i> ? (Karnal)	10	— (0)	Trace of purplish lilac (1)	Trace of pinkish lilac	Light perilla purple	Absent	Absent
	15	— (0)	Trace of dark hellobore red (2)	Vinaceous purple	Dark perilla purple	Thin	Do.
	20	Trace of purplish lilac (3)	Trace of purplish lilac (2)	Bishop's purple	Do.	Do.	Do.
	25	Trace of light purplish lilac (3)	White (3)	Light perilla purple	Dull violet black	Do.	Do.
	30	Trace of purplish lilac (2)	Trace of purplish lilac (2)	Perilla purple	Dark perilla purple	Do.	? A very thin layer
	35	Trace of light hyssop violet (3)	Trace of light hyssop violet (3)	Dark hyssop violet	Dark hyssop violet	Do.	Absent
F 57 Gram wdt organ-ism	10	— (0)	— (0)	Unchanged	Unchanged	Thin	Absent
	15	— (0)	White (1)	Do.	Do.	Do.	Do.
	20	White (1)	White (1)	Do.	Do.	Do.	Do.
	25	White (2)	White (1)	Do.	Do.	Do.	Do.
	30	White (1)	White (2)	Do.	Do.	Do.	Do.
	35	White (2)	White (1)	Do.	Do.	Do.	Do.

F 92 Gram wilt organ- ism.	10	— (0) .	White (1) .	Unchanged .	Unchanged .	Absent
	15	White (1) .	White (2) .	Do. .	Thin .	Do.
	20	White (2) .	White (1) .	Do. .	Do. .	Do.
	25	White (2) .	White (2) .	Do. .	Do. .	Do.
	30	White (2) .	White (2) .	Do. .	Do. .	Do.
	35	White (3) .	White (2) .	Do. .	Do. .	Do.
F 93 Gram wilt organ- ism.	10	— (0) .	— (0) .	Unchanged .	Unchanged .	Absent
	15	White (1) .	White (2) .	Do. .	Do. .	Do.
	20	White (2) .	White (3) .	Do. .	Thin .	Do.
	25	White (1) .	White (2) .	Do. .	Do. .	Do.
	30	White (2) .	White (2) .	Do. .	Do. .	Do.
	35	White (1) .	White (1) .	Do. .	Absent	Do.

Footnote—

1. * Coloured at the top of one tube only.

2. The figures following the colours in columns 3 and 4 refer to the abundance of aerial mycelium:
(0) Lacking; (1) Trace; (2) Moderate; (3) Abundant.

TABLE XI

Conidia and chlamydospores (Form of conidia and chlamydospores observed on the twenty-second and twenty-third days. Measurements made on the twenty-fourth and twenty-fifth days)

Culture	Temperature series, °C	Conidia in aerial mycelium	Conidia in pionotal layer	Chlamydospores
F 79 <i>F. bostrycoides</i>	20	Mainly single, but occasionally in false heads. Continuous, ovoid to spindle-shaped, hyaline. Abundant. 0-sept. 5.8μ long.	Pionnotes lacking	Absent
	35	Single. Continuous, ovoid to spindle-shaped, hyaline. Abundant 0-sept. 5.9μ long.	Ditto	Terminal and intercallary. Single. 1 and 2-celled. Smooth. Hyaline. Abundant.
F 80 <i>F. conglutinans</i> .	20	Single. Continuous, ovoid to spindle-shaped, hyaline. Few. 0-sept. 8.8μ long.	Ditto	Terminal and intercallary. Single and in chains. 1- and 2-celled. Smooth. Hyaline. Moderate numbers.
	35	Single. Continuous, ovoid to spindle-shaped, sometimes slightly curved, hyaline. Few 0-sept. 8.3μ long.	Ditto	Terminal and intercallary. Single and in chains. 1- and 2-celled. Smooth or slightly rough. Hyaline. Abundant.
F 81 <i>F. conglutinans</i> var. <i>betae</i> .	20	Single. Continuous and ovoid to spindle-shaped, hyaline, few. Rarely 1- or 3-septate and then spindle-shaped or slightly curved and bluntly tapering at both ends, with no foot-cell. Cell walls and septations indistinct. 0-sept. (96 per cent) 8.9μ , 1-sept. (4 per cent) 16.2μ long.	Ditto	Intercallary. Single. 1- and 2-celled. Smooth. Hyaline. Rare.

F 81 <i>F. congluticans</i> var. <i>betae</i> .	35	Spores absent	Pionnotes lacking	Terminal and intercallary. Single and in chains. 1- and 2-celled. Smooth. Hyaline. Moderate numbers.
F 82 <i>F. congluticans</i> var. <i>calis-tepha</i> .	20	Aerial mycelium had collapsed, so that the spores could not be distinguished from those borne in pionnotes.	0-6-sept., the continuous spores ovoid to spindle-shaped, the septate spores spindle-shaped or slightly curved and bluntly tapering at both ends, with no distinct foot-cell. Cell walls and septations indistinct. Sometimes constricted at septations. Granular. Many spores containing chlamydospores. 0-sept. (79 per cent) 8.8 μ , 1-sept. (12 per cent) 12.3 μ , 3-sept. (5 per cent) 31.6 μ , 4-sept. (2 per cent) 38.4 μ , 5-sept. (1 per cent) 51.0 μ , 6-sept. (1 per cent) 46.9 μ long.	Terminal and intercallary. Single. 1-celled. Smooth. Hyaline. Abundant.
	35	Aerial mycelium lacking	0-5-sept., the continuous spores ovoid to spindle-shaped, the septate spores spindle-shaped or slightly curved and bluntly tapering at both ends, with no distinct foot-cell. Cell walls and septations fairly distinct. Highly vacuolated. 0-sept. (68 per cent) 11.0 μ , 1-sept. (26 per cent) 18.1 μ , 2-sept. (4 per cent) 33.1 μ , 3-sept. (2 per cent) 36.5 μ long.	Difficult to observe method of bearing, owing to lack of mycelium, but apparently terminal and intercallary 1- and 2-celled. Smooth Hyaline. Abundant.

TABLE XI—*contd.*

Culture	Temperature series, °C	Conidia in aerial mycelium	Conidia in pionnotal layer	Chlamydo-spores
F 83 <i>F. orthoceras</i> .	20	Single. Continuous, rarely 1-sept., ovoid to spindle-shaped. Hyaline. Abundant. 0-sept. (96 per cent) 6.4 μ , 1-sept. (4 per cent) 12.1 μ long.	Pionnotes lacking	Terminal and intercallary. Single. 1- and 2-celled. Smooth. Hyaline. Few.
F 83 <i>F. orthoceras</i> .	35	Single. Continuous, ovoid to spindle-shaped, rarely 1-or 2-sept., then sometimes slightly curved, bluntly tapering at both ends, septations indistinct. Highly vacuolated. Abundant. 0-sept., 7.5 long.	Pionnotes lacking	Terminal and intercallary. Single. 1- and 2-celled. Smooth. Hyaline. Abundant.
F 84 <i>F. orthoceras</i> var. <i>apii</i> .	20	Single. Continuous or 1-3-septate. Continuous spores ovoid to spindle-shaped. Septate spores spindle-shaped, sometimes slightly curved, bluntly tapering at both ends. Septations indistinct. Hyaline or slightly granular. Moderately abundant.	Usually continuous, occasionally 1-3-sept., ovoid to spindle-shaped, the longer spores slightly curved, rounded at both ends, with indistinct walls and septations, granular or vacuolated, often constricted at the septations. 0-sept. (88 per cent) 8.3 μ , 1-sept. (12 per cent) 21.1 μ long.	Terminal. Single. 1-celled. Smooth. Hyaline. Rare.
	35	Single. usually continuous, occasionally 1-2-sept., ovoid to spindle-shaped, septate spores sometimes slightly	Pionnotes lacking	Terminal and intercallary. Single. 1- and 2-celled. Smooth. Hyaline. Few.

F 85 <i>F. orthoceras</i> var. <i>apii</i> f. 1.	curved, bluntly tapering at both ends, septations indistinct. Highly vacuolated. Spores slightly constricted at septations. Moderately abundant. 0-sept. (96 per cent), 10-8 μ , 1-sept. (4 per cent) 17-24 long.	Pionnotes lacking	Terminal and intercallary. Single. 1- and 2-celled. Smooth. Hyaline. Few.
20	Single. Continuous. Ovoid to spindle-shaped. Hyaline. Rare. 0-sept. 7-24 long.		
35	Spores absent	Pionnotes lacking	Terminal and intercallary. Single. 1- and 2-celled. Smooth. Hyaline. Abundant.
F 73 <i>F. orthoceras</i> var. <i>psii</i> .	Spores absent	Pionnotes lacking	Terminal and intercallary. Single. 1-celled. Smooth. Hyaline. Moderate numbers.
20			
35	Single. Continuous. Ovoid to spindle-shaped. Hyaline, rare. 0-sept. (5 spores only) 5-6 μ long.	Ditto	Terminal and intercallary. Single. 1- and 2-celled. Some spores smooth, some very rough. Hyaline. Abundant.
F 86 <i>F. orthoceras</i> var. <i>longius</i> .	Single. Continuous, ovoid to spindle-shaped, hyaline, abundant.	0-3-sept., ovoid to spindle-shaped, the longer spores sometimes slightly curved, rounded at both ends, thin-walled and very faintly septate. Granular. 0-sept. (88 per cent) 12-9 μ , 1-sept. (10 per cent) 17-5 μ , 2-sept. (2 per cent) 19-7 μ long.	Absent.
20			

TABLE XI—*contd.*

Culture	Temperature series, °C	Conidia in aerial mycelium	Conidia in pionotal layer	Chlamydospores
F 87, <i>F. angustum</i> .	35	Single. Continuous or 1-sept., ovoid to spindle-shaped, sometimes slightly curved, with fairly distinct septations. Somewhat granular. Abundant.	Continuous or 1-sept., ovoid to spindle-shaped, sometimes slightly curved, rounded at both ends, thin-walled, hyaline. 0-sept. (90 per cent) 11.0 μ , 1-sept. (10 per cent) 18.2 μ long.	Terminal. Single. 1-celled. Smooth. Hyaline. Rare.
	20	Usually single, occasionally in false heads. Continuous, ovoid to spindle-shaped, hyaline, abundant. 0-sept. 7.1 μ long.	Pionotes lacking	Terminal and intercalary. Single. 1-celled. Smooth. Hyaline. Abundant.
	35	Single, continuous, ovoid to spindle-shaped, rarely 1-sept. with constrictions at the septa, septations indistinct. Granular. Abundant. 0-sept. 5.6 μ long.	Iditto	Terminal and intercalary. Single and in chains. 1- and 2-celled. Smooth. Hyaline. Abundant.
F 74 <i>F. lini</i> .	20	Single, continuous or 1-sept., ovoid to spindle-shaped. Septations indistinct. Hyaline. Abundant.	Continuous to 3-sept., the continuous spores ovoid to spindle-shaped, the septate spores spindle-shaped and often slightly curved, tapering bluntly at both ends, cell walls and septations rather indistinct, hyaline. 0-sept. (79 per cent) 10.6 μ ,	Terminal and intercalary. Single. 1- and 2-celled. Smooth. Hyaline. Few.

F 20 <i>F. lini</i> ? (Karnal).	35	Single, continuous and ovoid to spindle-shaped, rarely 0.3 sept. and gently tapering to blunt point at both ends, straight, septations indistinct. Hyaline. Abundant. 0-sept. 9.4 μ long.	Pionnotes lacking	1-sept. (14 per cent) 19.0 μ , 2-sept. (1 per cent) 28.9 μ , 3-sept. (4 per cent) 46.3 μ , 4-sept. (1 per cent) 62.9 μ , 5-sept. (1 per cent) 68.0 μ long.	Terminal. Smooth. Single. Hyaline. Abundant.
	20	Single, continuous, rarely 1-sept., ovoid to spindle-shaped, hyaline, moderately abundant. 0-sept. (98 per cent) 7.0 μ , 1-sept. (2 per cent) 16.5 μ long.	Ditto		Intercalary. Single. Hyaline. Smooth. Rare.
	35	Single, continuous and ovoid to spindle-shaped or 1- or 2-sept. and spindle-shaped or slightly curved and tapering to blunt point at both ends. Cell walls and septations fairly distinct. Hyaline. Abundant. 0-sept. (90 per cent) 9.4 μ , 1-sept. (10 per cent) 18.7 μ long.	Ditto		Terminal and intercalary. Single. 1- and 2-celled. Smooth Hyaline. Abundant.
F 57 Gram wilt organism.	20	Single, continuous, ovoid to spindle-shaped, hyaline, Rare. 0-sept. 9.5 μ long.	Ditto		Absent

TABLE XI—*concl.*

Culture	Tempera- ture series, °C	Conidia in aerial mycelium	Conidia in pionotal layer.	Chlamydo-spores
F 92 Gram wilt organism.	35	Single. Usually continuous, rarely 1-sept., ovoid to spindle-shaped, sometimes slightly curved. Hyaline. few. 0-sept. 9.1μ long.	Pionnotes lacking . . .	Absent. (Six days later, on re-examination, two single, 1-celled, smooth, hyaline spores were found).
	20	Single, continuous, ovoid to spindle-shaped. Few.	Ditto . . .	Intercalary. Single. 1-celled. Smooth. Hyaline. rare.
	35	Single, continuous or 1-sept., ovoid to spindle-shaped, sometimes slightly curved. Septations fairly distinct. Hyaline. Few. 0-sept. (96 per cent) 9.6μ, 1-sept. (4 per cent) 16.5μ long.	Ditto . . .	Terminal and intercalary. Single. 1- and 2-celled. Smooth. Hyaline. Moderately abundant.
F 93 Gram wilt organism.	20	Single. Continuous, rarely 1-sept., ovoid to spindle-shaped, hyaline, few. 0-sept. (98 per cent) 10.6μ, 1-sept. (2 per cent) 20.4μ long.	Ditto . . .	Absent

35	Single. Usually continuous and ovoid to spindle-shaped, occasionally 1-3-sept. and spindle-shaped or slightly curved and tapering to blunt points at both ends. Cell walls and septations fairly distinct. Hyaline. Abundant. 0-sept. (98 per cent) 9-9 μ , 1-sept. (2 per cent) 25-24 long.	Ditto	.	.	Terminal and intercalary. Single, 1-celled. Hyaline. Moderately abundant.
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The main conclusions from this experiment can be summarized as follows :-

- (1) Except with *F. orthoceras* var. *apii* f. 1 the only effect of temperature on colour was a slightly more rapid production of pigment at the higher temperatures, at which the fungi grew more quickly, and a tendency towards production of a slightly more violet hue. All the cultures showing the purple pigment turned red with two per cent hydrochloric acid and violet or blue with two per cent potassium hydroxide.

F 85 (*F. orthoceras* var. *apii* f. 1) produced no pigment at 10°, 15°, 30° and 35°C. It produced purple aerial mycelium and dark plumbeous discoloration of the substrate at 20°C, and at 25°C one tube was not pigmented while the duplicate had a small patch of pigment at the thin end of the slant. (This particular tube formed the material for experiment 3).

- (2) In general, the cultures showed the greatest amount of aerial mycelium at 20°, 25° and 30°C. Very little was produced at 10°C, and there was generally a falling off at 35°C. Casual observation suggested that this character was correlated with the rate of linear growth at the various temperatures, but no detailed records on this point were kept.
- (3) All the cultures except *F. bostrycoides* had a thin plectenchymatous stroma, and in most cases this was present at all temperatures except 10°C.
- (4) A thin pionnotal layer of spores was formed by *F. conglutinans* var. *callistephi*. Several cultures, namely *F. orthoceras* var. *apii*, *F. orthoceras* var. *longius*, *F. angustum* and *F. lini* had a very thin layer of spores almost too scanty to deserve the name 'pionnotal', though 'pionnotes' are referred to in Table XI. There was no regular relationship between temperature and the production of this very thin layer of spores.
- (5) Abundance of spores in the aerial mycelium varied considerably with the different species, but a difference in temperature of 15°C (20° as compared with 35°C) had no appreciable effect. Spores were rare with *F. orthoceras* var. *apii* f. 1 and *F. orthoceras* var. *psi*. The three other varieties of this species had abundant or moderately abundant spores in the aerial mycelium.
- (6) Most of the cultures produced only continuous or 1-septate, ovoid, spindle-shaped or slightly curved spores in the aerial mycelium. The only ones producing 3-septate spores in the aerial mycelium were *F. conglutinans* var. *betae*, *F. orthoceras* var. *apii*, *F. lini*, and the gram wilt fungus F 93.
- (7) The so-called pionnotes had more septate spores, and in the case of *F. conglutinans* var. *callistephi* occasional 4-5-6-septate spores were found. Even here, however, the non-septate spores totalled 68 per cent of the whole.

- (8) The effect of temperature on either the number of septations or the length of spores of a given number of septations was slight. Averaging the lengths of all comparable sets of 0-septate spores in the aerial mycelium, namely *F. bostrycoides*, *F. conglutinans*, *F. orthoceras*, *F. angustum*, *F. lini*, and the two gram wilt fungi F 57 and F 93, the mean lengths at 20°C are 7.9 μ and at 35°C, 8.0 μ , a negligible difference. Insufficient 3-septate spores were available for comparison. *F. bostrycoides* had unusually small 0-septate spores, and *F. orthoceras* var. *longius* had rather large ones in the thin superficial or pionnotal layer. If the spores in the pionnotal layers of *F. orthoceras* var. *longius* may strictly be compared with those in the aerial mycelium of *F. orthoceras*, the differences between these two varieties of one species is greater than the difference between any two species except *F. bostrycoides*.
- (9) In most cases temperature had a marked effect on chlamydospore production, 35°C being favourable and 20°C unfavourable. The difference was most marked in the cultures of *F. bostrycoides*, *F. conglutinans* var. *apii* f. 1, *F. lini*, and the gram wilt fungi F 92 and F 93.

PRODUCTION OF PIGMENT BY *F. ORTHOCERAS* VAR. *APII* F 1

It is recorded in Table X that at the temperature 20°C culture F 85 (*F. orthoceras* var. *apii* f. 1 Wr. and Reink. = *F. apii* var. *pallidum* Nelson and Sherbakoff) produced a purplish lilac hue in the aerial mycelium and dark plumbeous in the substrate, and at 25°C also one tube showed a trace of colour at the shallow end of the agar. Since Nelson, Coons and Cochran [1937] say of this fungus 'Mycelium and substratum always colourless' this colour production attracted particular attention.

From the tube showing a trace of colour, six transfers were made from the coloured portion and six from the white portion, on fresh oatmeal agar on the twenty-seventh day, and three fresh tubes were kept at 25°C. They were numbered, respectively, 1-6 W (from white portion) and 1-6 C (from coloured portion). When ten days old the colours were examined. The results are given in Table XII.

It is seen that the aerial mycelium of all cultures produced a mixture of white and purplish lilac, that the substrate was colourless at the bottom or deep end of all the slants, but that the top or shallow end of the substrate was coloured in one tube only in the case of the cultures from the colourless portion of the parent and in four tubes in the case of the cultures from the coloured portion of the parent tube. An attempt was then made to obtain by further sub-culturing, cultures which produced entirely colourless and others which produced entirely coloured colonies. In order to do this, transfers were again made, this time from the top and bottom portions of the slants of tubes 1W and 1C. Four cultures of each were made. This was done when the tubes were ten days old. These tubes were labelled as follows, and placed at 25° C:—

1 W T (inoculated from top of tube 1 W)

1 W B (inoculated from bottom of tube 1 W)

1 C T (inoculated from top of tube 1 C)

1 C B (inoculated from bottom of tube 1 C).

TABLE XII

Colour production in F. orthoceras var. apii f. 1 Wr. and Reink. (=F. api var. pallidum Nelson and Sherbakoff) after ten days

Tube No.	Nature of parent portion of culture	Colour of aerial mycelium		Colour of surface of substrate	
		Top of slant	Bottom of slant	Top of slant	Bottom of slant
1 W	Colourless . .	White and purplish lilac.	Light mouse gray	Unchanged .	Unchanged
2 W	Do. . .	Do. . .	Do. . .	Do. . .	Do.
3 W	Do. . .	Do. . .	Do. . .	Do. . .	Do.
4 W	Do. . .	Do. . .	Do. . .	Purplish lilac .	Do.
5 W	Do. . .	Do. . .	Do. . .	Unchanged .	Do.
6 W	Do. . .	Do. . .	Do. . .	Do. . .	Do.
1 C	Coloured . .	Do. . .	Do. . .	Purplish gray .	Do.
2 C	Do. . .	Do. . .	Do. . .	Unchanged .	Do.
3 C	Do. . .	Do. . .	Do. . .	Purplish gray .	Do.
4 C	Do. . .	Do. . .	Do. . .	Unchanged .	Do.
5 C	Do. . .	Do. . .	Do. . .	Purplish gray .	Do.
6 C	Do. . .	Do. . .	Do. . .	Do. . .	Do.

The same day transfers were also made from the original pair of cultures grown at 25°C which were by now 37 days old. These cultures also were prepared in quadruplicate, and they were marked thus :—

O W T (inoculated from top of original colourless tube)

O W B (inoculated from bottom of original colourless tube)

O C T (inoculated from top of original partially coloured tube)

O C B (inoculated from bottom of original partially coloured tube, but from the colourless portion)

In these eight series the replicate tubes were labelled a, b, c and d. Their colours were observed on the 11th and again on the 21st days. The colours observed on the twenty-first day are recorded in Table XIII.

At the conclusion of this experiment the tubes showing colours were divided into two groups, one of which had two per cent hydrochloric acid added, and the other 2 per cent potassium hydroxide. In all cases the purple or gray hues became red with hydrochloric acid and violet or blue with potassium hydroxide.

TABLE XIII

Colour production of F. orthoceras var. apii f. 1 Wr. and Reink. (=F. apii var. pallidum Nelson and Sherbakoff) in first and second sub-cultures from white and coloured tubes, after twenty-one days.

First sub-culture

Culture*	Colour of aerial mycelium		Colour of surface of substrate	
	Top of tube	Bottom of tube	Top of tube	Bottom of tube
O W T a	White and argyle purple	White and light mouse gray	Unchanged . .	Unchanged
b	White and dark vinaceous gray	White and deep mouse gray	Do. . .	Do.
c	Do. . .	Do. . .	Do. . .	Do.
d	Do. . .	Do. . .	Do. . .	Do.
O W B a	White and vinaceous gray	White and argyle purple and light mouse gray	Do. . .	Ivory yellow
b	White . . .	White and mouse gray	Do. . .	Unchanged
c	White and purplish lilac	Do. . .	Do. . .	Ivory yellow
d	White and dark vinaceous gray	White and light mouse gray	Do. . .	Unchanged
O C T a	White . . .	Do. . .	Do. . .	Ivory yellow
b	White and dark vinaceous gray	Do. . .	Deep quaker drab .	Do.
c	White . . .	Do. . .	Unchanged . .	Do.
d	White and dark vinaceous gray	Do. . .	Do. . .	Unchanged
O C B a	Do. . .	Do. . .	Do. . .	Do.
b	Do. . .	Do. . .	Do. . .	Do.
c	Do. . .	Do. . .	Do. . .	Do.
d	Do. . .	Do. . .	Do. . .	Do.

*O W T—from top of original colourless tube.

O W B—from bottom of original colourless tube.

O C T—from top (coloured portion) of original coloured tube.

O C B—from bottom (colourless portion) of original coloured tube.

TABLE XIII—*contd.**Second sub-culture*

Culture†	Colour of aerial mycelium			Colour of surface of substrate	
	Top of tube		Bottom of tube	Top of tube	Bottom of tube
1 W T a	White lilac	and purplish	White and light mouse gray	Pale vinaceous lilac.	Ivory yellow
b	Do.	.	Do.	Do.	Do.
c	Do.	.	Do.	Do.	Do.
d	Do.	.	White	Deep purplish vinaceous	Do.
1 W B a	Do.	.	White and light mouse gray	Unchanged	Do.
b	Do.	.	Do.	Vinaceous lavender	Do.
c	White	.	Do.	Unchanged	Do.
d	White lilac	and purplish	Do.	Do.	Do.
1 C T a	White	.	Do.	Do.	Do.
b	White lilac	and purplish	White and deep mouse gray	Light vinaceous lilac	Do.
c	White	.	White and light mouse gray	Unchanged	Do.
d	White lilac	and purplish	White and deep mouse gray	Light vinaceous lilac	Do.
1 C B a	White	.	White and light mouse gray	Unchanged	Do.
b	White lilac	and purplish	Do.	Do.	Do.
c	Do.	.	Do.	Light vinaceous lilac	Do.
d	Do.	.	Do.	Unchanged	Do.

†1 W T—inoculated from top of colourless tube originating from colourless tube.

1 W B—inoculated from bottom of colourless tube originating from colourless tube.

1 C T—inoculated from top of coloured tube originating from coloured tube.

1 C B—inoculated from bottom of coloured tube originating from coloured tube.

The results of this experiment appear to indicate that both the original coloured tube and the originally colourless one consisted of a mixture of two strains, one capable of producing a pigment with acid and alkali reactions typically those of the sub-section *Orthocera*, the other unable to do so. There was no indication that sub-culturing from the upper portions of the slant, where the colour was intense, would yield an intensely pigmented culture. If the coloured strain occurred as a saltant it must have done so at a stage previous to the inoculation of Experiment 1.

It is interesting to record that one of the most intensely coloured cultures was retained in the stock culture collection and has been sub-cultured four or five times. Its power to produce the pigment has dwindled but not entirely disappeared.

INFLUENCE OF ASPARAGINE ON THE KEY CHARACTERS

The purpose of this experiment was to find whether a variation in the asparagine content of a synthetic medium influenced the septation of spores, colour of mycelium and substrate, and other characters as found by Brown [1925] in *Fusarium fructigenum*.

The following five cultures were used :—

F 74. *F. lini* Bolley :

In previous experiments this culture showed a distinct tendency to produce a purplish lilac colour in the aerial mycelium and a deep purple, lilac or violet hue in the substrate, varying presumably with the acidity or alkalinity of the medium. It also produced a thin layer of spores on the agar surface, resembling pionnotes. In nature, according to Wollenweber and Reinking [1935] it sometimes produces sporodochia, and in the form of the conidia is a bridge between *F. orthoceras* and *F. oxysporum*. It produced abundant chlamydospores at 35°C, few at 20°C.

F 85. *F. orthoceras* var. *apii* f. 1 Wr. and Reink. (= *F. apii* var. *pallidum* Nelson and Sherbakoff) :

This culture behaved peculiarly in regard to colour production, sometimes producing a purplish lilac colour in the aerial mycelium and perilla purple or dark plumbeous in the substrate, at other times having white aerial mycelium and producing no colour in the stroma. It produced no pionnotes and even non-septate small conidia in the aerial mycelium were few. It produced abundant chlamydospores at 35°C, few at 20°C.

F 57. Gram wilt fungus :

This culture produced no colour, and very rarely produced septate spores. Small conidia were few in the aerial mycelium. Production of chlamydospores was very rare at 35°C, and none were found at 20°C.

F 92. Gram wilt fungus :

Resembled F. 57 except in the moderate production of chlamydospores at 35°C and occasionally at 20°C.

F 93. Gram wilt fungus :

This culture, like F 57 and F 92, was colourless, but it produced some long 3-septate spores and the smaller spores in the aerial mycelium were more abundant than in either F 57 or F 92. Chlamydospores were moderately abundant at 35°C, though absent at 20°C.

The media used in the experiment were as follows :—

1. Glucose 2 gm.
 K_2PO_4 1.25 gm.
 $MgSO_4 \cdot 7H_2O$ 0.75 gm.
 Agar 15 gm.
 Water 1 litre
2. Medium 1 plus 0.05 gm. asparagine

3. Medium 1 plus 0.10 gm. asparagine
4. Medium 1 plus 0.20 gm. asparagine
5. Medium 1 plus 0.50 gm. asparagine
6. Medium 1 plus 1.00 gm. asparagine
7. Medium 1 plus 2.00 gm. asparagine
8. Medium 1 plus 4.00 gm. asparagine
9. Oatmeal agar, using 100 gm. oatmeal per litre, steamed at 60°C for one hour, strained through muslin, sterilized at 10 lb. pressure for 45 minutes (20 gm. agar)
10. Plain agar (20 gm. per litre).

Four tubes of each medium were inoculated with each fungus from plain agar cultures, except in the case of *F* 93, tubes of which were inoculated from 3 per cent oatmeal agar because the growth on plain agar was unsatisfactory. The cultures were grown at 25°C. The replicates were labelled *a*, *b*, *c* and *d*, and these designations are used in Tables XV and XVI which summarize the observations on spore forms. The colours noted on the nineteenth day are recorded in Table XIV.

A most surprising feature of this experiment was the complete failure of the fungi to produce any pigment whatsoever on any of the synthetic media, in spite of excellent growth with an abundant covering of aerial mycelium. These media are known to be less satisfactory for colour production than similar media to which potato starch is added, but complete failure to produce colour by such a deeply pigmented culture as *F. lini* was not expected. Starch was deliberately omitted because Brown [1925] did not find it suitable for studying sporulation in the feebly sporing strains of *F. fructigenum* (= *F. ateritium*).

As was the case in experiment 2 on oatmeal agar, *F. orthoceras* var. *apii* *f*. 1 formed practically no septate spores. Its behaviour on other media was irregular. For instance, few spores were found on medium 5 containing 0.5 gm. of asparagine, but spores were present in abundance on media 4 and 6, containing 0.2 and 1.0 gm. of asparagine respectively. Spores were absent in medium 8, containing 4.0 gm. of asparagine. *F. lini* produced non-septate spores in media 1 (with rare exception), 2, 4 and 6. It produced 0-3-septate spores in two tubes of medium 3, one tube of medium 5, two tubes of medium 7 and one tube of medium 8. It produced only continuous spores on oatmeal agar and plain agar. Only medium 5 gave sufficient 3-septate spores of *F. lini* for a reliable average, and they measured $33.2\mu \times 4.1\mu$.

The lack of agreement between replicate tubes as regards chlamydospore production was most striking.

DISCUSSION

It is not often possible in any genus of fungi containing a large number of species to name any single variable character the exact measurement of which will determine the species. It is in fact fairly widely accepted that a one-character difference, unless it is very great indeed, can hardly be considered to warrant anything higher than varietal rank. Moreover, when the particular character concerned is highly variable and responds readily to environmental changes, its value is considerably reduced.

TABLE XIV
Colour production on synthetic media and on plain agar and oatmeal agar after 19 days

Medium	<i>F. lini</i> F 74		<i>F. orthoceras</i> var. <i>ajacii</i> f. 1 F 85		Gram wilt fungus F 57		Gram wilt fungus F 92		Gram wilt fungus F 93	
	Surface of Substrate	Mycelium	Surface of Substrate	Mycelium	Surface of Substrate	Mycelium	Surface of Substrate	Mycelium	Surface of Substrate	Mycelium
1	Unchanged	Lacking	Unchanged	Lacking	Unchanged	Lacking	Unchanged	Lacking	Unchanged	Lacking
2	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.
3	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.
4	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.
5	Do.	White	Do.	Do.	Do.	White	Do.	White	Do.	White
6	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.
7	Do.	Do.	Do.	White	Do.	Do.	Do.	Do.	Do.	Do.
8	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Do.	Lacking
9 (oatmeal)	Dark hyssop violet	White with trace of hyssop violet	White with dark mouse gray at bottom of one tube	Do.	White	Do.	White	Do.	White	White
10 (Plain agar)	Unchanged	Lacking	Unchanged	Lacking	Unchanged	Lacking	Unchanged	Lacking	Unchanged	Lacking

TABLE XV

Conidia obtained from surface scrapings of synthetic media, plain agar and oatmeal agar after 20—21 days
(Replicate tubes designated a, b, c, d)

Medium	<i>F. lini</i> F 74	<i>F. orthoceras</i> var. <i>apii</i> f. 1 F 85	Gram wilt fungus F 57	Gram wilt fungus F 92	Gram wilt fungus F 93
1	Continuous (rarely ovoid to spindle-shaped, hyaline, abundant. a, b, c, d.	Continuous, ovoid to spindle-shaped, hyaline, moderately abundant.	Continuous, ovoid to spindle-shaped, hyaline, abundant. a, b.	0-2-sept., ovoid to spindle-shaped or slightly curved and bluntly pointed at both ends. Hyaline. Moderately abundant. a, b.	Continuous, ovoid to spindle-shaped, hyaline, few. Showing a tendency to cluster into false heads. a, b.
2	Continuous, ovoid to spindle-shaped, hyaline, abundant. a, b, c, d.	Continuous, ovoid to spindle-shaped, hyaline, moderately abundant.	Continuous or 1-sept., ovoid to spindle-shaped, hyaline, abundant. a, b.	0-3-sept., ovoid to spindle-shaped when continuous, curved and tapering to a blunt point at both ends when septate. Hyaline. Continuous spores abundant. Septate distinct. Septate spores few. a, b, c, d. 3-sept. $36.5 \times 3.6\mu$	Continuous, ovoid to spindle-shaped, hyaline, abundant. a, b.
3	0-3-sept., continuous spores ovoid to spindle-shaped, septate spores spindle-shaped or slightly curved, bluntly pointed at both ends, hyaline, septations fairly distinct. Septate spores in a and b only, in considerable numbers.	Continuous, ovoid to spindle-shaped, hyaline. Moderately abundant.	Continuous or 1-sept. (one 3-sept. spore observed), ovoid to spindle-shaped, hyaline, abundant.	0-3-sept., ovoid to spindle-shaped when continuous, curved and tapering to a blunt point at both ends when septate. Hyaline. Continuous spores abundant. Septa distinct. Septate spores few. a, b, c, d. 3-sept. $34.7 \times 3.5\mu$	Continuous, ovoid to spindle-shaped hyaline, abundant. a, b.
4	Continuous, ovoid to spindle-shaped, some spores vacuolated but mostly hyaline. Abundant. a, b, c, d.	Continuous ovoid to spindle-shaped, hyaline, abundant.	0-3-sept. (one 4-sept. spore observed), the continuous spores ovoid to spindle-shaped and abundant, the septate ones spindle-shaped or	0-3-sept., ovoid to spindle-shaped or slightly curved and tapering to a blunt point at both ends when septate. Hyaline.	Continuous, ovoid to spindle-shaped, hyaline, abundant. a, b.

5	In tubes <i>a</i> , <i>b</i> , <i>d</i> , continuous, ovoid to spindle-shaped, vacuolate, abundant. In <i>c</i> long spores in great abundance, 0-3 sept., cylindrical or slightly curved, tapering to blunt point at both ends, highly vacuolated, without foot cell. Total of 1195 spores examined consisted of 92.2 per cent 0-sept., 4.5 per cent 1-sept., 0.9 per cent 2-sept., and 2.4 per cent 3-sept. 3-sept. $33.2\mu \times 4.1\mu$.	Continuous, ovoid to spindle-shaped, few.	Continuous or 1-sept., ovoid to spindle-shaped, hyaline, abundant. <i>a</i> , <i>b</i> .	Continuous spores abundant, septate spores rare. Septa distinct. <i>a</i> , <i>b</i> , <i>c</i> , <i>d</i> . 3-sept. $33.8 \times 3.7\mu$ (average of six spores only).	Continuous, ovoid to spindle-shaped, abundant in <i>b</i> , <i>c</i> and <i>d</i> . In <i>a</i> , some (rare) 3-sept. spores, spindle-shaped or slightly curved and tapering to a blunt point at both ends. Septations very indistinct. Vacuolated. 3-sept. $27.8 \times 3.4\mu$.
6	Continuous, ovoid to spindle-shaped, abundant, vacuolated. <i>a</i> , <i>b</i> , <i>c</i> , <i>d</i> .	Continuous, ovoid to spindle-shaped, hyaline, moderately abundant.	Continuous or 1-sept. (one 3-sept. spore observed), ovoid to spindle-shaped or slightly curved, hyaline, abundant. <i>a</i> , <i>b</i> .	0-3-sept., ovoid to spindle-shaped when continuous, spindle-shaped or slightly curved and tapering to a blunt point at both ends when septate. Hyaline. Continuous spores abundant, septate spores few. Septa distinct. <i>b</i> . 3-sept. $34.3 \times 3.7\mu$.	Continuous, ovoid to spindle-shaped, hyaline, moderately abundant. <i>a</i> , <i>b</i> .
7	In <i>a</i> and <i>c</i> , continuous, ovoid to spindle-shaped, vacuolated, abundant. In <i>b</i> and <i>d</i> , 3-sept. spores also seen, spindle-shaped or slightly curved, tapering to blunt point at both ends, highly vacuolated, few.	Continuous, ovoid to spindle-shaped, hyaline, few.	Continuous or 1-sept., ovoid to spindle-shaped or slightly curved, hyaline, abundant. <i>a</i> , <i>b</i> .	0-3-sept., ovoid to spindle-shaped when continuous, spindle-shaped or slightly curved and tapering to a blunt point at both ends when septate. Hyaline. Continuous spores abundant, septate spores few. Septa distinct. <i>a</i> , <i>b</i> , <i>c</i> , <i>d</i> . 3-sept. $31.1 \times 3.8\mu$.	Continuous, or 1-septate ovoid to spindle-shaped, hyaline, abundant. <i>a</i> , <i>b</i> .
8	In <i>b</i> , <i>c</i> and <i>d</i> , continuous, ovoid to spindle-shaped, vacuolated, abundant. In <i>a</i> , 3-sept. spores also seen, spindle-shaped or slightly curved, tapering to blunt point at both ends, highly vacuolated, few.	Absent	Continuous, ovoid to spindle-shaped, hyaline, abundant. <i>a</i> , <i>b</i> .	0-3-sept., ovoid to spindle-shaped when continuous, spindle-shaped or slightly curved and tapering to a blunt point at both ends when septate. Hyaline. Continuous spores abundant, septate spores rare. Septa distinct. <i>a</i> , <i>b</i> , <i>c</i> , <i>d</i> . 3-sept. $32.7 \times 4.0\mu$ (mean of 4 spores only).	Continuous or 1-sept., ovoid to spindle-shaped, hyaline, abundant. <i>a</i> , <i>b</i> .

TABLE XV—*contd.*

Medium	<i>F. lini</i> F 74	<i>F. orthoceras</i> var. <i>apiis</i> f. 1 F 85	Gram wilt fungus F 75	Gram wilt fungus F 92	Gram wilt fungus F 93
9 (Oatmeal agar)	Continuous, ovoid to spindle-shaped, vacuolated, abundant <i>a, b, c, d.</i>	Continuous (rarely 1-sept.) ovoid to spindle-shaped, sometimes slightly curved, hyaline, abundant.	Continuous, ovoid to spindle-shaped, hyaline, abundant. <i>a, b.</i>	0-3-sept., ovoid to spindle-shaped when continuous, curved and tapering to a blunt point at both ends when septate. Hyaline. Continuous spores abundant, septate spores also fairly abundant. Septa distinct. <i>a, b.</i> Total of 686 spores examined consisted of 92.7 per cent 0-sept., 4.8 per cent 1-sept., 0.6 per cent 2-sept. and 2.0 per cent 3-sept. 3-sept. 30 × 3.7 μ.	Continuous or 1-sept., ovoid to spindle-shaped, hyaline moderately abundant. <i>a, b.</i>
10 (Plain agar)	Continuous, ovoid to spindle-shaped, vacuolated, few. <i>a, b, c, d.</i> (Growth very poor)	Absent (Growth very poor).	Continuous, ovoid to spindle-shaped, hyaline, few. <i>a, b.</i> (Growth very poor)	0-1-sept., ovoid to spindle-shaped, hyaline, few. (Growth very poor)	Continuous, ovoid to spindle-shaped, hyaline, few. <i>a, b.</i> (Growth very poor).

TABLE XVI

Chlamydospore production on synthetic media, plain agar and oatmeal agar after 21 days (Replicate tubes designated a, b, c, d)

Medium	F 74	F 85	F 57	F 92	F 93
1	Terminal, single, smooth, hyaline. Rare. <i>a, b, c, d</i>	Absent	Absent	Absent	Absent
2	Absent <i>a, b, c, d</i>	Terminal and intercalary, single, 1-celled, hyaline, smooth, few.	Terminal, single, 1-celled, smooth, hyaline, few.	Terminal and intercalary, single, 1-celled, smooth, hyaline, rare.	Do.
3	Terminal and intercalary, single, 1-celled. Mostly smooth and hyaline, but sometimes warty and brown. Abundant in <i>a</i> and <i>b</i> . Absent in <i>c</i> and <i>d</i>	Absent	Terminal and intercalary, single, 1-celled, smooth, hyaline, abundant.	Terminal, single, 1-celled, smooth, hyaline, rare.	Do.
4	Absent <i>a, b, c, d</i>	Absent	Terminal and intercalary, single, 1-celled, smooth, hyaline, abundant.	Terminal, single, 1-celled, smooth, hyaline, rare.	Do.
5	Absent <i>a, b, c, d</i>	Terminal, single, 1-celled, smooth, few	Terminal and intercalary, single, 1-celled, smooth, hyaline, abundant.	Terminal and intercalary, single, 1-celled, smooth, hyaline, rare.	Do.
6	Terminal and intercalary, single, 1-celled smooth, hyaline. Moderately abundant in <i>b</i> , rare in <i>d</i> , absent in <i>c</i> and <i>c</i> .	Terminal, single, 1-celled, smooth, hyaline, few	Terminal and intercalary, single, 1-celled, smooth, hyaline, abundant.	Terminal and intercalary, single, 1-celled smooth, hyaline, moderate numbers.	Do.
7	Absent <i>a, b, c, d</i>	Absent	Terminal and intercalary, single, 1-celled, smooth, hyaline, few.	Absent <i>a, b</i>	Do.
8	Absent <i>a, b, c, d</i>	Absent	Terminal and intercalary, single, 1-celled, smooth, hyaline, few.	Absent <i>a, b</i>	Do.
9	Absent <i>a, b, c, d</i>	Terminal, single, 1-celled, smooth, hyaline, rare	Terminal and intercalary, single, 1-celled, smooth, hyaline, moderately abundant.	Terminal and intercalary, single, 1-celled, smooth, hyaline, abundant.	Do.
10	Absent <i>a, b, c, d</i>	Absent	Terminal and intercalary, single, 1-celled, smooth, hyaline, abundant.	Terminal and intercalary, single, 1-celled, smooth, hyaline, abundant.	Do.

The section *Elegans* has a number of characteristics which are said to typify the section. The more important of these are :—

- (1) Presence of abundant, mostly one-celled, ovoid to spindle-shaped, small conidia, not borne in chains.
- (2) Presence of terminal and intercalary chlamydospores.
- (3) Delicate walls and septations in the conidia.

The species in the sub-section *Orthocera* have the additional characteristics of slenderness of the spores, which are almost straight or spindle-shaped, with a papillate or very slightly foot-celled base. They are without, or at most with traces of sporodochia.

The experiments described here have indicated that the authentic cultures studied produce in many cases abundant small conidia of the kind described. *F. conglutinans* and *F. conglutinans* var. *apii* f. 1 rarely produced any at all. Steamed rice was in many cases unsatisfactory for their production, but the remaining media used, namely oatmeal agar, potato cylinders, two per cent potato dextrose agar and five per cent potato dextrose agar, were about equally satisfactory.

All the cultures were found to produce chlamydospores, but they were found to vary to an extraordinary degree in this respect. *F. orthoceras* var. *longuis*, and the gram wilt fungus F 57 produced chlamydospores very rarely. There was a striking difference between abundance of chlamydospores at 20° C and at 35°C, the latter usually greatly enhancing their production.

The conidia had delicate cell-walls and septations, and this characteristic did not seem to alter with the medium or the temperature. It seems to be a characteristic of the group.

Owing to the fact that some of the cultures failed to produce an appreciable number of 3-septate spores, the ratios of length to breadth were not determined, but in all cases they were spindle-shaped or only slightly curved, and they had no typical foot-celled base. These again appear to be characteristics of the group.

Provided these fungi were grown at 35°C they could readily be identified as *Orthocera-Fusaria* with the possible exception of *F. orthoceras* var. *apii* f. 1, *F. orthoceras* var. *longuis* and the gram wilt fungus F 57, for which a large number of tubes might have to be examined before reaching a final decision.

On the whole, the description of the sub-section given by Wollenweber and Reinking seems to be satisfactory and it covers the degree of variation exhibited by the different members.

The distinction between the various species depends, as stated previously, on the following major characteristics :—

- (1) Presence or absence of pionnotes.
- (2) Type of conidiophores.
- (3) Colour of stroma.
- (4) Type of plectenchyma-erumpent or smooth.
- (5) Sizes of conidia.
- (6) Pathogenicity.

The type of conidiophore branching is used only for distinguishing *F. bostrycoides*. As stated, no bostrycoid branching could be found; the method of branching of the conidiophores in this species appeared identical with all the other species of the group.

Pathogenicity is a very positive characteristic, but it has not been practical to determine its variability.

According to Wollenweber and Reinking sclerotia may or may not occur in the sub-section. It is a noteworthy fact, however, that of the 12 representatives they describe, sclerotia are stated to be absent in four and no mention is made of them in seven others. The only case where they are mentioned is in *F. lini*, a species which they consider to be, as regards form of macroconidia, a bridge between *F. orthoceras* and *F. oxysporum*. The latter fungus produces sclerotia. No doubt it is very convenient to leave this trying species, *F. lini*, out of the key. It is questionable whether any of the fungi produced any structure which could genuinely be called a 'stroma'. The nearest approach was a plectenchymatous layer, and the fungi differed little in their formation of this. The term 'stroma' has been used in this paper to describe the thin superficial, fleshy layer which forms on the surface of all moderately nutritious agars. It has not been found to be erumpent.

We are left with three variable characters which might be used as an aid to identification, namely, presence or absence of pionnotes, colour of 'stroma', and sizes of conidia. What are they worth?

The only culture definitely producing pionnotes was *F. conglutinans* var. *callistephi*. Four other species, namely *F. orthoceras* var. *apii*, *F. orthoceras* var. *longuis*, *F. angustum* and *F. lini* had very thin superficial layers of spores which were barely entitled to the name 'pionnotes', and could best be described in Wollenweber and Reinking's words 'Konidienschleime von geringer Ausdehnung'. It so happens that *F. conglutinans* var. *callistephi* is placed by Wollenweber and Reinking in the group with pionnotes typically absent, though in their detailed description they say that the conidia of this variety are more or less copiously scattered about, in exceptional cases covering the substrate as a faint, thin transitory pionnotes.

The colours of 'stroma' or of surface of the substrate lend themselves better than many characters to accurate description because they can be compared with well-known colour standards. It has been clearly shown that there were three main groups in the cultures studied, based on pigmentation. These groups may be compared with the colours described by Wollenweber and Reinking:—

Culture	Colour of 'stroma' or surface of substrate	Colour of stroma according to Wollenweber and Reinking
<i>F. orthoceras</i> var. <i>pisi</i>	Blue-brown-hued . . .	Reddish ochre to chestnut-brown
<i>F. bostrycoides</i> . . .	Purple hued, changing to red in HCl and blue or violet in KOH	Brownish white, then palm-green or violet
<i>F. orthoceras</i> , . .	Ditto . . .	Pale, flesh-coloured, green-flecked, purple-red-violet (becoming blue in alkali)

Culture	Colour of 'stroma' or surface of substrate	Colour of stroma according to Wollenweber and Reinking
<i>F. orthoceras</i> var. <i>apii</i>	Purple hued, changing to red in HCl and blue or violet in KOH	Pale, flesh-coloured, greeno flecked, purple-red-violet (becoming blue in alkali)
<i>F. orthoceras</i> var. <i>longuis</i>	Ditto	Ditto.
<i>F. angustum</i>	Ditto	Rose to purple-red (becoming blue in alkali)
<i>F. lini</i>	Ditto	Various coloured, clear, brownish white, flesh coloured, greenish, rose to red (in alkali violet or blue)
<i>F. orthoceras</i> var. <i>apii</i> f. 1	Usually non-pigmented, sometimes variously grayish-purple	Pale, not becoming reddish-violet on rice mush nor blue in alkali
<i>F. conglutinans</i>	Non-pigmented	Pale, white, then brownish to rosy white
<i>F. conglutinans</i> var. <i>betae</i>	Non-pigmented	Pale, white, then brownish to rosy white
<i>F. conglutinans</i> var. <i>callistephi</i>	Ditto	Pale, white, then yellowish, brownish to rosy-white, in exceptional cases with traces of grayish lilac colour
F 57 Gram wilt organism	Usually non-pigmented, sometimes variously grayish-purple
F 92 Gram wilt organism	Ditto
F 93 Gram wilt organism	Ditto

The case of *F. orthoceras* var. *apii* f. 1 throws considerable light on the whole question of pigmentation. According to Nelson, Coons and Cochran [1937] this fungus is supposed to produce no pigmentation. It is clearly shown that it may produce it under certain conditions, and the pigment shows the usual acid and alkali reactions.

Apart from *F. orthoceras* var. *pisi* there is only one significant pigment produced, namely the purple pigment becoming red in hydrochloric acid and blue or violet in potassium hydroxide. That certain isolates of *F. orthoceras* var. *pisi* may also produce this pigment is suggested by the work of Snyder [1933].

The remaining characteristic which lends itself to accurate measurement is spore size. It has been shown that as regards the 0-septate spores the variation within so-called species is as great as or greater than the variation between species, and the character is therefore of no value for specific determination.

Sizes of 3-septate spores could not in many cases be determined owing to the fact that some cultures produced few or no such spores. In the case of *F. lini* comparisons may be made with the figures of Wollenweber and Reinking :—

Culture	Measurements by Wollenweber and Reinking	Author's measurements
<i>F. lini</i>	35 × 4 (21–41 × 2.5–4.5) .	On Brown's medium with 0.05 per cent asparagine (25 spores), 33.2 × 4.1 μ (18.7–51.0 × 3.4–5.8) On 3 per cent oatmeal agar at 20° C (4 spores only) 46.3 μ in length (39.1–52.7)

It is clear that in *F. lini* the variation is much greater than indicated by Wollenweber and Reinking.

The three gram-wilt organisms F 57, F 92 and F 93, though alike in lack of colour, show marked differences in regard to spore production. F 57 and F 93 produce very few 3-septate spores, but when they do produce them they are not appreciably different in size, and the range of variation of the means is not greater than that of *F. lini*. Spores of similar septations are alike in the three isolates as regards form. Duplicate tubes of these and of other cultures vary greatly in the ratios of continuous to septate spores. All the three gram-wilt organisms can produce chlamydospores, but in abundance of these they differ.

We are forced to the conclusion that the cultures of *Orthocera-Fusaria* maintained at Baarn cannot, with one or two exceptions, be recognized from the descriptions given by Wollenweber and Reinking. The key simply falls to the ground when used with authentic cultures.

According to Wollenweber and Reinking, *Fusarium lini* fruits better than the other species of this section, and in many isolations even produces occasional sporodochia, and therefore belongs to a transitional form with the other groups. This view is strengthened by their statement that the macroconidia are, in form, a bridge between *F. orthoceras* and *F. oxysporum*. The original description of *F. lini* given by Bolley [1901] definitely refers to sporodochia with typically 4-celled conidia. If we are to include the other fungi in this species it means that we are practically obliged to regard them as only sub-normal material of *Oxysporum*—or *Constrictum-Fusaria*, a procedure which Wollenweber and Reinking [1935] give reasons for not adopting.

F. angustum, according to the original description by Sherbakoff [1915] differs from *F. orthoceras* and *F. conglutinans*, in its spore form narrowly tapering at the ends and sometimes anguiform. The lack of septate spores in the experiments described here have made it impossible to study the reliability of this character. A study of the original description makes one wonder why this fungus was ever placed in the sub-section *Orthocera*. The shape of spores pictured by Sherbakoff, with their narrowly tapering ends and their distinct

foot-cells, together with the high length : breadth ratio, would seem to eliminate it completely from this sub-section, and bring it in line with the *Constrictum-Fusaria*. Wollenweber and Reinking [1935] have retained it in the sub-section *Orthoceras* in spite of a length : breadth ratio of 13 : 1, which is quite outside the limits of the group.

F. bostrycoides has not in any of the observations made shown the bostrycoid branching by which it is supposed to be recognizable and from which it has taken its name. It has, however, a very distinct tendency to produce conidia in false heads. For the time being it seems advisable to retain this species, though eventually it may have to undergo union with one of the others.

We are left with *Fusarium orthoceras* and *F. conglutinans*. It has been suggested by Wollenweber and Reinking [1935] that these two might be united, but it has not been done because the fungi grew somewhat differently. In the experiments described here, however, all distinctions have completely broken down. In many respects the differences between varieties within one of these species are greater than the differences between the two species, and all efforts have failed to reveal a characteristic difference between the so-called species. It seems that we are fully justified in uniting the two under the name *Fusarium orthoceras* App. and Wollenweber.

Fusarium conglutinans was the name given by Wollenweber [1913] for the fungus causing wilt disease of cabbage. The description was as follows :—

‘Differs from *F. orthoceras* in the absence of a wine-red colour on rice, which is a striking character of typical species of the section *Elegans*. Vascular parasite, cause of wilt disease of *Brassica oleracea* var. *capitata* (proved by Erwin F. Smith, L. R. Jones and L. L. Harter) in the United States of America.’

It will be seen that the difference lies only in pigment production. Yet Wollenweber and Reinking mention a rosy-white colour in *F. conglutinans* and *F. conglutinans* var. *callistephi*, and conversely, as we have seen, non-pigmented strains are found in varieties of *F. orthoceras*. If this were accepted as the significant difference between the two species, one wonders why Wollenweber and Reinking placed *F. apii* var. *pallens* of Nelson and Cochran (*F. apii* var. *pallidum* Nelson and Sherbakoff) as a variety of *F. orthoceras* and not of *F. conglutinans*. The answer, of course, is that there is no fundamental difference between the species.

Fortunately, *F. orthoceras* is one of the species of *Fusarium* which at the start had an adequate description [Appel and Wollenweber, 1910]. This certainly cannot be said of *F. conglutinans*. The original description of *F. orthoceras* covers the description of *F. conglutinans* and its varieties *betæ* and *callistephi*, apart from the characters of pathogenicity. It is therefore proposed to rename these three fungi as follows :—

Fusarium orthoceras App. et Wollr. var. *conglutinans* n.c.

Syn. *F. conglutinans* Wr.

Morphologically indistinguishable from the fundamental species. Cause of a vascular wilt disease of *Brassica oleracea* in North America (U. S. A.).

Fusarium orthoceras App. et Wollr. var. *betæ* n.c.

Syn. *F. conglutinans* Wr. var. *betæ* Stewart.

Morphologically indistinguishable from the fundamental species. Cause of a seedling blight of *Beta vulgaris* in North America (U. S. A.).

Fusarium orthoceras App. et Wollr. var. *callistephi* n.c.

Syn. *F. conglutinans* var. *majus* Wr.

F. conglutinans Wr. var. *callistephi* Beach.

Morphologically indistinguishable from the fundamental species. Cause of a vascular wilt disease of *Callistephus chinensis* in most countries where this plant is grown.

The gram wilt fungi are considered to comprise one variety :—

Fusarium orthoceras App. et Wr. var. *ciceri* n. var. Morphologically indistinguishable from the fundamental species. Cause of a vascular wilt disease of *Cicer arietinum* in India.

In the opinion of the author, the decision of Nelson, Coons and Cochran [1937] to change the names of *Fusarium orthoceras* var. *apii* Woll. et Rkg. and *F. orthoceras* var. *apii* f. 1 Woll. and Rkg. to *Fusarium apii* and *F. apii* var. *pallidum* respectively is unfortunate. The decision was based not on experimental evidence that the fungi concerned are morphologically different from *F. orthoceras* but on the opinion that pathogenic considerations should be a major criterion in distinguishing species—‘The most important differential character is the distinct host relationship and it is chiefly on this basis that the segregation is made.’ When Linford in 1928 created the variety *pisi* of *F. orthoceras* based on the ability of this fungus to cause a wilt of *Pisum* he laid the foundations of a nomenclatorial system for these fungi which was already of proved worth in *Puccinia graminis* with its varieties. Linford’s procedure appears to be the one most likely to avoid confusion.

SUMMARY

(1) Morphological and cultural studies have been made of eleven of the twelve species, varieties or physiologic forms of *Fusarium* of the sub-section *Orthocera*, using cultures supplied by the Centraalbureau voor Schimmelcultures, Baarn. *Fusarium conglutinans* var. *citrinum* Wr. (*F. citrinum* Wr.), the remaining species, could not be obtained. Included in the experiments were three fungi able to cause wilt of gram (*Cicer arietinum*).

(2) The cultures vary in ability to produce spores in the aerial mycelium, to produce septate spores, to produce chlamydospores, to produce pigments etc.

(3) In respect of pigment production, the cultures fall in three groups :—

(i) Producing blue or brown pigments, unaffected by addition of hydrochloric acid or potassium hydroxide.

(ii) Producing a purple pigment, becoming red in hydrochloric acid and blue or violet in potassium hydroxide.

(iii) Non-pigmented.

While all cultures of *F. conglutinans* failed to produce pigments, varieties or forms of *F. orthoceras* fell within all three groups. Steamed rice is an excellent medium for studying pigment production.

(iv) Production of aerial mycelium, and of a so-called ‘stroma’, and size of non-septate spores are of no value in identifying these species.

(v) Pigment production appeared not to be appreciably influenced by temperature of growth except with *F. orthoceras* var. *apii* f. 1 Woll., a fungus reported to be non-pigmented, but producing the purple pigment in some cultures at 20° and 25° C.

(vi) The only culture producing typical pionnotes was *F. conglutinans* var. *callistephi* which is placed by Wollenweber and Reinking in the group with pionnotes typically absent.

(vii) The effect of temperature on either the number of septations or the length of conidia was slight, but a temperature of 35° C was markedly more favourable for chlamydospore production than one of 20° C.

(viii) An experiment on the effect of asparagine in a synthetic medium on the septation or length of spores were inconclusive and it was found that replicate cultures often gave entirely different results.

(ix) The results are discussed in detail and reasons are given for regarding *Fusarium conglutinans* as a synonym of *F. orthoceras*, which should be divided up into varieties based on major pathogenic capabilities. Reasons are given for not uniting *F. orthoceras* with the earlier *F. lini* or with *F. bostrycoides* or *F. angustum*. It is not clear, in fact, why *F. angustum* should be placed in the sub-section *Orthocera* at all.

(x) *F. conglutinans*, *F. conglutinans* var. *betae*, and *F. conglutinans* var. *callistephi*, become varieties of *F. orthoceras*, and the new variety *F. orthoceras* var. *ciceri* is proposed.

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CYTOLOGICAL STUDIES IN *GOSSYPIMUM*

I. CHROMOSOME BEHAVIOUR IN THE INTERSPECIFIC HYBRID *G. ARBOREUM* × *G. STOCKSII*

BY

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(With 13 text-figures)

INTRODUCTION

WITH the object of transferring the drought-resistant qualities of the Asiatic wild cotton *G. Stocksii* Mast. to the strains of rain fed cultivated cottons of southern India, hybridization between them was attempted by the Cotton Specialist, Coimbatore. The hybrids between the local Karunganni strain K1 (*G. arboreum* L. var. *neglectum* Watt forma *indica* H. & G.) ($2n=26$) and *G. Stocksii* Mast. ($2n=26$), proved to be completely sterile. A cytological examination of the hybrid was undertaken to discover the causes of its sterility.

Meiosis in similar hybrids between *G. Stocksii* Mast. and other types of *G. arboreum* L. have been examined by Skovsted [1937].

MATERIAL AND METHODS

All the seeds and plant material required for the investigation were kindly supplied by Rao Bahadur V. Ramanatha Ayyar, Cotton Specialist, Coimbatore. Flower buds for the study of meiosis were collected from plants which were grown at the Cotton Breeding Station and at the Imperial Sugarcane Breeding Station, Coimbatore.

Flower buds were fixed in acetic alcohol for 24 hours, washed in 90 per cent alcohol and then dehydrated and infiltrated by the chloroform method [La Cour, 1931]. A thickness of 18 to 20 μ was found necessary for anther sections. Slides stained in gentian violet gave good results.

Temporary aceto-carmin mounts of either fresh or fixed material were made according to the method suggested by Bellings [1926] and used for microscopic examination and drawings. Such temporary mounts, after being ringed, were quite suitable for critical examination and fit for use for nearly five to seven days. A good number of drawings of meiotic stages reproduced in this paper were made from such temporary mounts.

MEIOSIS IN THE PARENTS

Chromosome pairing and chiasma behaviour have been studied in both the parents at diplotene, diakinesis and metaphase stages and the data are

presented in Table I and II (Appendix). To ensure accuracy in determinations observations were made on 78 bivalents at each stage in both the species, confining observations only to uncut nuclei showing complete complement of chromosomes.

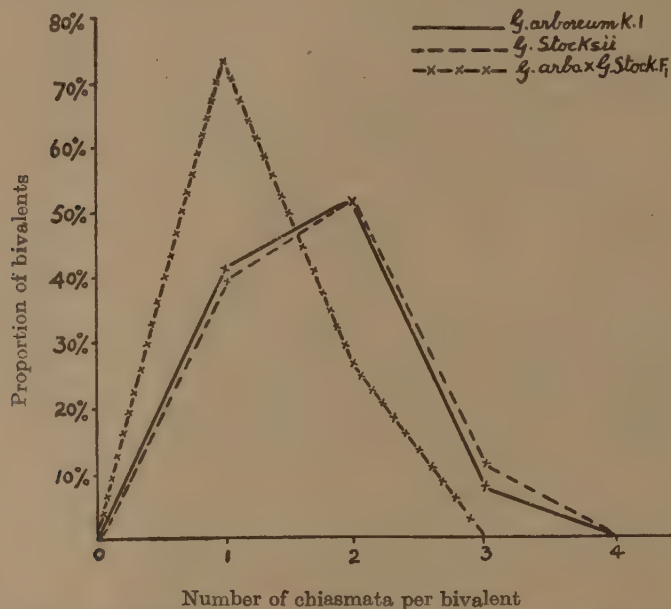


FIG. 1. Curves showing the chiasma frequency per bivalent in the two parents and the hybrid

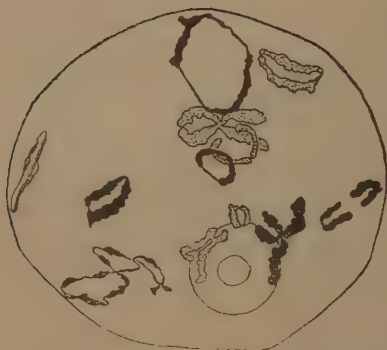


FIG. 2. Diplotene stage in *G. arboreum* K1 ($\times 1000$)

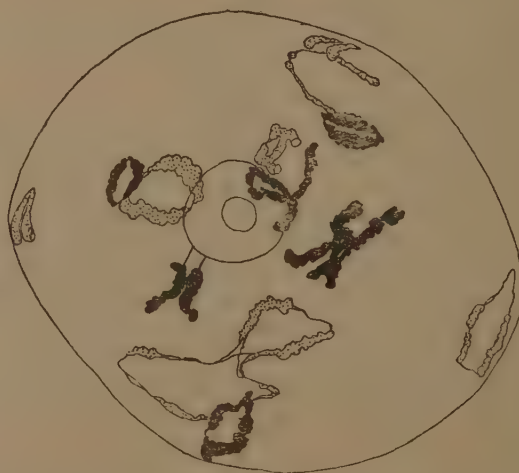


FIG. 3. Diplotene stage in *G. Stocksii*, ($\times 2100$)

The following observations have been recorded :

The number of chiasmata in each bivalent varied from one to three in both the species, two being most common. V—, X—, O—, and 8-shaped bivalents were observed (Figs. 2 and 3). Except for slight variations from nucleus to nucleus, the general appearance and configurations of the bivalents at the same stage were nearly the same in both the species. The mean chiasmata per bivalent at diplotene was nearly 1.7 in both the species. Terminalization of chiasmata was incomplete in both the species. There was increase in the coefficient of terminalization from diplotene to metaphase (Tables I and II and Fig. 1). Between diplotene and diakinesis stages there was very little reduction of the mean chiasmata per bivalent, whereas there was a definite reduction between diakinesis and metaphase. Differential contraction of chromosomes has been observed at diplotene in both the species, some bivalents having shortened and thickened more rapidly than others (Figs. 2 and 3).

The first and second meiotic divisions have been found to be quite normal giving rise to normal tetrads and pollen grains.

MEIOSIS IN THE HYBRID

In the sterile F_1 hybrid between the two species also, observations were made in the stages from diploptene to metaphase. Just as in the case of the parents, six complete nuclei showing all the bivalent and univalent chromosomes were selected for observations at each stage. The number of bivalents in a nucleus was seen to vary from five to nine, the average number per nucleus being nearly seven. The rest of the chromosomes remained as univalents (Figs. 5 and 6). The number of chiasmata in a bivalent varied from one to two only, the majority having only a single chiasma (Figs. 4 and 1). The mean chiasmata per bivalent at diplotene was, therefore, only 1.3 which was considerably less than that in the parents. This indicated that the affinity even among the pairing chromosomes in the hybrid was less than that in the parents. V—, X—, O—, and 8—shaped bilavents could be seen. Terminalization of chiasmata in the bivalents was incomplete as in the parents (Fig. 1 and Table III), but the reduction of the mean chiasmata per bivalent due to terminalization was gradual. The terminalization coefficients at the three stages were lower than those in the parents (Fig. 1 and Table III).

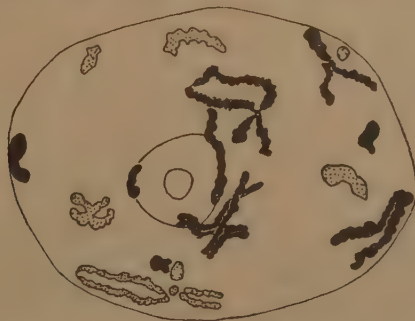


FIG. 4. Diplotene stage in *G. arboreum* K1 \times *G. Stocksii* F_1 ($\times 1000$)

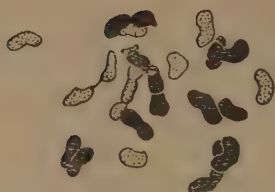


FIG. 5. Meiotic metaphase stage in the hybrid showing bivalents and univalents. ($\times 2100$)



FIG. 6. Anaphase (meiotic) in the hybrid showing arc-shaped chromatid threads ($\times 1000$)

In the large number of pollen mother cells examined, no configurations higher than bivalents could be seen. The numbers of bivalents were determined in 24 pollen mother cells which gave an average of 7.13 per pollen mother cell. The details are given in the following table.

Frequency of different combinations of chromosome configurations

No. of different combinations found	Chromosome configurations		No. of pollen mother cells
	Univalents	Bivalents	
1	16	5	1
2	14	6	4
3	12	7	11
4	10	8	7
5	8	9	1

Total of univalents	.	.	.	282
Total of bivalents	.	.	.	171
Mean number of univalents	.	.	.	11.75
Mean number of bivalents	.	.	.	7.13

As is usual in many sterile interspecific hybrids, at metaphase, the bivalents and the univalents were seen scattered about at random in the cytoplasm (Fig. 5). This irregular arrangement was due to the fact that all the chromosomes have not reached the equatorial plate and arranged themselves in the normal compact manner. Although most of the bivalents arranged themselves at the equatorial plate, very often a few of them were seen away from the plate, scattered about in the cytoplasm along with the univalents (Figs. 6 and 7). When the paired chromosomes began to separate and an anaphase spindle was formed, the univalents were seen distributed at random on the spindle (Fig. 8). The fate of the univalents during this division was decided by their position in relation to the separating bivalents. Those situated far away from the equator moved with the daughter bivalents passing to the nearest pole, while those situated in or near about the equatorial plate scattered in the cytoplasm, either in groups or singly, without undergoing any division at first metaphase (i.e. predivision). Such irregular movements of the univalents relative to those of the bivalents that divided gave rise to various kinds of abnormalities in the grouping of the chromosomes at the end of the first division. Besides the two main groups of chromosomes, those that were extruded from the daughter nuclei into the cytoplasm were generally seen scattered singly or in groups of varying numbers (Fig. 11). Occasionally, however, certain pollen-mother-cells showed two distinct compact second metaphase plates (Fig. 12) with unequal numbers of chromosomes.



FIG. 7. Anaphase (meiotic) in the hybrid showing separating bivalents connected by chromatic threads. Notice the non-parallel chromatic threads. ($\times 1000$)



FIG. 8. Anaphase (meiotic) in the hybrid showing a regular spindle. ($\times 2100$)

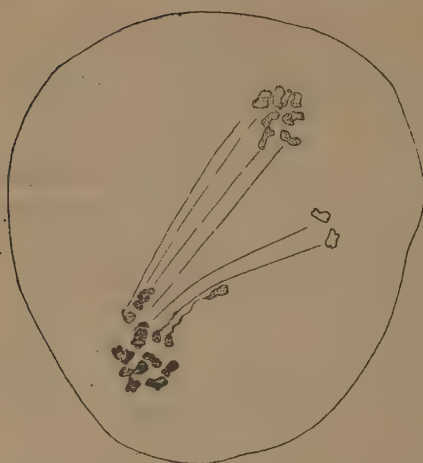


FIG. 9. Pollen mother cells showing anaphases of Division I in the hybrid. Notice the division of one of the poles into two. ($\times 1000$)

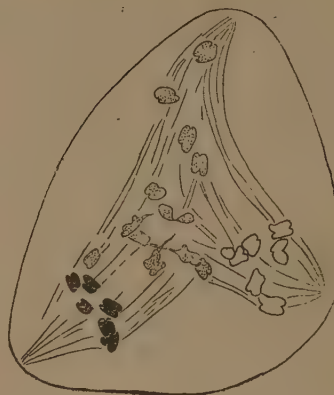
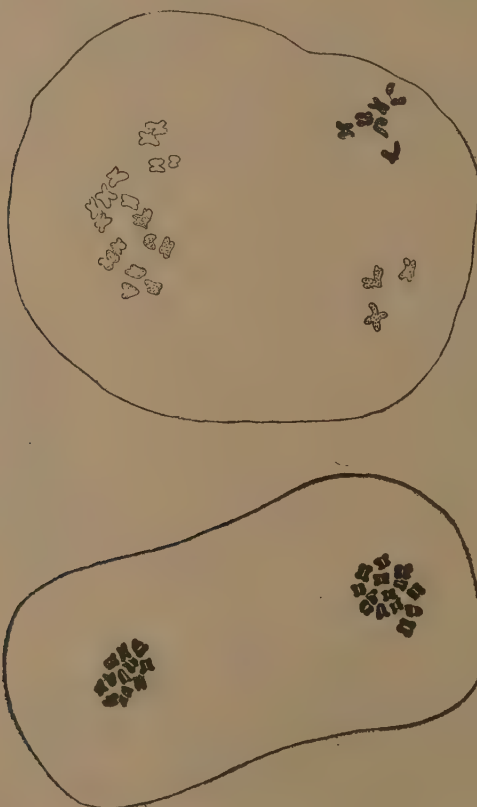


FIG. 10. Pollen mother cell of the hybrid showing a tripolar spindle at Division I. ($\times 2100$)



FIGS. 11 and 12. Pollen mother cells of the hybrid showing second metaphase plates. ($\times 1000$)

The abnormal anaphase movements of the chromosomes mentioned above have been found to be associated with various irregularities of the first division spindle. In many pollen mother cells, although the univalents were scattered at random at anaphase, the spindle was straight and bipolar (Fig. 8) like the normal spindles of the parents. In most of the pollen mother cells, the separating daughter bivalents, although sufficiently far away from each other, appeared to be connected by slender chromatin threads (Figs. 6 and 7). The unwinding spirals of the chromatin threads, as a result of the pulling force could be clearly seen. The bivalents were evidently under a constant stress of the pulling force, which naturally caused pulling out of the non-separated parts of the chromatin threads. Some of the cells showed the chromatin threads connecting the separating bivalents, bent in the form of arcs (Fig. 6). The direction of the pull indicated the orientation of the spindle threads and showed that the spindle threads also were of the same arc-shape, as if both the poles were situated on the same side of the cell. Tripolar spindles were also observed in some pollen-mother-cells (Fig. 10). In others a division has been found to occur at one of the two poles (Fig. 9). This has given rise to three main second metaphase plates with a few chromosomes extruded into the cytoplasm either singly or in groups.

As has been shown above, these abnormalities of the first division left the chromosomes scattered in several groups or singly (Fig. 11), and each of these groups or solitary chromosomes formed a second metaphase plate. The number of such plates were seen to vary from two to six. The chromosomes of these second metaphase plates were either univalents, daughter bivalents, or even undivided bivalents, which were all capable of division at this stage. The second division spindles were, therefore, seen to be normal (Fig. 13) in all the pollen-mother-cell examined. The second metaphase plates which contained a small number of chromosomes divided, giving rise to micronuclei and micropollen grains. Occasionally, a plate which contained only a single chromosome, remained undivided (Fig. 13) giving rise to a micropollen grain with a single diad chromosome. Thus it is seen that the pollen grains produced were without the normal complement of chromosomes. In a total of 194 pollen-mother-cells examined, the number of pollen grains produced by a pollen-mother-cell have been found to vary from three to eleven.

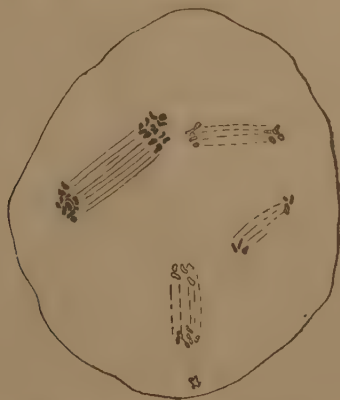


FIG. 13. A pollen mother cell of the hybrid showing second division spindles. See the solitary undivided diad chromosome. ($\times 1250$)

DISCUSSION

Causes of sterility

It is evident from the observations made that sterility in this hybrid is due mainly to incomplete pairing of chromosomes at meiosis, as was observed by Skovsted [1935, 1937] in similar interspecific hybrids between these two species and other hybrids among Asiatic cottons. Such failure of pairing is generally caused by lack of homology of various degrees between the chromosomes of the two species involved. Absence of homology in the present instances may be attributed to loss of homology of the chromosomes of the two species as a result of geographical isolation for long periods and their consequent independent evolution, as suggested also by Skovsted [1935, 1937]. The degree of incompatibility brought about in the genomes of the two species by such divergent evolutions was vividly shown by the abnormal meiotic behaviour of the chromosomes in the hybrid. As mentioned above, these abnormalities have been found to be closely associated with irregularities in the development of the spindle. According to Darlington and Thomas [1937], a normal spindle is developed by the coordinate action of two agents in the cell, one outside the nucleus and the other inside the nucleus and, therefore, a defective spindle may arise from 'faults' in the 'mutual adjustment' of the two agents. The extra nuclear agents responsible for spindle development in plants, according to these authors, correspond in function to the centrosomes found in animals and some lower plants, but are of a different character in that they may 'be supposed to exist as diffused particles which coalesce or congregate at the moment when spindle poles are normally formed'. On the other hand the internal agent is the coordinated action of the centromeres of the dividing chromosomes. 'It seems that to prevent the spindle stretching and bending it is necessary not only to have paired chromosomes, but also to have them there at the right time' [Darlington, 1937]. Therefore, it follows that variations in the irregularities of the spindle may depend also on the varying numbers of paired chromosomes present. It has been shown that in many of the pollen mother cells of the present hybrid the spindle was straight and bipolar (Fig. 8) like the normal spindles of the parents. It was quite likely that in such cells, the number of bivalents were proportionately high and the majority of them reached the equatorial region in time, formed a regular plate and then separated more or less at the same time, thus controlling the development of the regular spindle. In cells where the number of paired chromosomes ready to divide were comparatively at a minimum, i.e. five or six, the spindle had a tendency to bend in the form of arcs (Fig. 6). This was perhaps due to lack of pairing or to 'the spindle developing too early in relation to the chromosomes' [Darlington, 1937]. Such bent spindles have been observed in many other organisms also e.g. *Drosophila* [Koller, 1934], *Impatiens* [Smith, 1935]. It was mentioned that in a large majority of the pollen-mother-cells, at first anaphase, the separating bivalents, although sufficiently far away from each other, appeared to be still connected by chromatin threads. This non-separation of the chromatin threads for a long time, in spite of the influence of the spindle on the anaphase movement of the daughter bivalents, may probably have been due to the fact that the

spindle began its action in the process of division before the bivalents were ready to separate, and that may have naturally caused pulling out of the non-separated parts of the chromatin threads. The formation of tripolar spindles was also among the abnormalities observed (Figs. 9 and 11). This may be attributed to the 'congregation or coalescence' of the pole determining material (external agent) in more than two regions, as shown by Darlington and Thomas [1937] in a *Festuca-Lolium* derivative. Even this 'congregation or coalescence' at a certain pole may sometimes be rather diffuse as was indicated by the non-parallel spindle threads in certain pollen-mother-cells (Fig. 7). All these irregularities of cell division at once indicate a probable fault in the timing adjustment of the two agents responsible for spindle development. Besides loss of homology between their chromosomes, each of the two parents of this hybrid may have, by their divergent evolution, acquired different characteristics and different timings in the various stages of cell division. When they are brought together in hybridity naturally a certain amount of disagreement may occur in the timings of the various stages of cell division as indicated in the observations made. This kind of timing unbalance in the movements of the chromosomes of the hybrid was observed even at the prophase stages. It has been mentioned already that in the parents themselves, there was a certain amount of differential condensation of the chromosomes at the diplotene stage and that such differences could not be observed at diakinesis and later stages. In the hybrid, however, differential condensation of the chromosomes was very marked and persisted even up to the diakinesis stage. It may be due to this timing unbalance among the chromosomes of the hybrid that some of the paired chromosomes at metaphase were not ready to come to the equatorial plate for division (Figs. 6 and 7), and it may be the same disharmony that enhanced the abnormalities of the spindle.

Inter-relationship

It has been shown that out of the 13 pairs of chromosomes of the two parents, *G. arboreum*-K1 and *G. Stocksii* Mast., only about seven pairs (i.e. nearly half) are homologous, while the other six chromosomes appear to be non-homologous. Skovsted's work [1937] has indicated that chromosome homology of some degree exists in all the interspecific hybrids studied. This has led him to think that all the cottons concerned are of monophyletic origin. From the point of view of chromosome homology he has further shown that *G. Stocksii* Mast. is less related to the Old World cottons than the other wild cottons, *G. Sturtii* F. M. and *G. anomalum* Wawra and Peyr. When hybrids between Old World cultivated cottons and the wild cottons, *G. anomalum* Wawra and Peyr. and *G. Sturtii* F. M., show bivalents varying from 9.5 to 11.85, the hybrids between *G. Stocksii* Mast. and the Old World cultivated cottons show bivalents varying from only 3.2 to 7.05 [Skovsted, 1937]. Moreover, the former set of hybrids show a few trivalents and quadrivalents, whereas the latter set do not seem to show any configurations higher than bivalents. In the present hybrid also the average number of bivalents per pollen-mother-cell does not exceed 7.13, and there appears to be no evidence of any auto-syndesis taking place. These evidences seem to indicate a distinct difference between *G. Stocksii* Mast. and the other two Old World wild cottons in their

relationships with the Old World cultivated cottons. In this connexion special attention may be drawn to the fact that in all the hybrids of which *G. Stocksii* Mast. is one of the parents, on an average, not more than about seven pairs of chromosomes seem to be homologous. It may, therefore, follow that the remaining (i.e. six pairs) chromosomes of this species, which do not pair with the chromosomes of the other Old World cottons, might have originated from a source entirely different from the source from which the corresponding set of chromosomes in the other Old World cottons have arisen. Davie [1933] and Skovsted [1937], basing their evidence on secondary pairing of chromosomes, have suggested, that the 26 chromosomes condition of the diploid cottons represent a secondary condition derived from a lower ancestral number. The morphological distinction between the two pairs of satellited chromosomes of the somatic complements of these two species, pointed out by the author [Abraham 1940], also lend support to this view. If this is the real state of affairs, then it may be suggested, in view of the evidence from chromosome pairing given above, that the 26 chromosomes conditions of *G. Stocksii* Mast. was derived, from a lower number of six or seven chromosomes, by a method different from that by which the other species have derived it. Further cytological studies on the inter-relationships among Old World cottons might clear up this issue.

SUMMARY

1. In the two cottons, *G. arboreum* L. var. *neglectum* Watt, forma *indica* H. & G.—strain K1 and *G. Stocksii* Mast., the first and second meiotic divisions were found to be normal.
2. The mean chiasmata per bivalent at diplotene was nearly 1.7 in both.
3. The reduction of the mean chiasmata per bivalent due to terminalization between diplotene and diakinesis was slight (0.03 in *arboreum* and nil in *Stocksii*), whereas there was a considerable reduction between diakinesis and metaphase (0.08 in *arboreum* and 0.10 in *Stocksii*).
4. In the hybrid chromosome pairing was incomplete, the number of bivalents in a nucleus varying from five to nine with an average of 7.13.
5. The mean chiasmata per bivalent at diplotene was only 1.3 and the reduction of the mean chiasmata per bivalent due to terminalization was gradual.
6. The anaphase movements of the chromosomes and the development of the first division spindle in the hybrid were found to be highly irregular.
7. Besides normal spindles, tripolar and bent spindles have been observed.
8. Sterility in the hybrid is found to be the result of the irregularities of cell division, caused by incomplete pairing of chromosomes at meiosis.
9. It is likely that irregularities in cell division may be caused also by differences in the timings of the various stages of cell division acquired by the parent species as a result of their divergent evolution.
10. In all crosses between *G. Stocksii* Mast. and other Old World cottons so far examined, not more than about seven chromosomes of the two species were found to be homologous. This suggests that the remaining six chromosomes of *G. Stocksii* Mast. have had an origin entirely different from that of the corresponding set in other Old World cottons.

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Appendix

TABLE I

Summary of observations of chiasma behaviour

G. arboreum—K1

	Diplotene	Diakinesis	Metaphase
1 Nos. of chiasmata			
Total	130	128	122
Interstitial	29	19	15
Terminal	101	109	107
Terminalization coefficient . .	0.777	0.825	0.877

Appendix—contd.

TABLE I—contd.

	Diplotene	Diakinesis	Metaphase
<i>1a Nos. of chiasmata per bivalent</i>			
Total	1.66	1.64	1.56
Interstitial	0.37	0.24	0.19
Terminal	1.29	1.40	1.37
<i>2 Nos. of bivalents with</i>			
1-chiasma	32	30	34
per cent	41.0	38.4	43.6
2-chiasmata	40	46	44
per cent	51.3	59.0	56.4
3-chiasmata	6	2	..
per cent	7.7	2.6	..
<i>3 Terminalization coefficient</i>			
1-chiasma	0.656	0.667	0.618
2-chiasmata	0.875	0.935	0.977
3-chiasmata	0.556	0.500	..

Appendix—*contd.*

TABLE II

*Summary of observations of chiasma behaviour**G. Stocksii*

	Diplotene	Diakinesis	Metaphase
1 Nos. of chiasmata			
Total	132	132	124
Interstitial	33	24	23
Terminal	99	108	101
Terminalization coefficient . .	0.750	0.818	0.815
1a Nos. of chiasmata per bivalent			
Total	1.69	1.69	1.59
Interstitial	0.42	0.31	0.29
Terminal	1.27	1.38	1.30
2 Nos. of bivalents with			
1-chiasma	31	28	32
per cent	39.7	35.9	41.0
2-chiasmata	40	46	46
per cent	51.3	59.0	59.0
3-chiasmata	7	4	..
per cent	9.0	5.2	..
3 Terminilization coefficient			
1-chiasma	0.613	0.714	0.656
2-chiasmata	0.863	0.870	0.869
3-chiasmata	0.524	0.667	..

Appendix—contd.

TABLE III

*Summary of observations of chiasma behaviour**G. arboreum—K1 × G. Stocksii—F₁*

	Diplotene	Diakinesis	Metaphase
1 Nos. of chiasmata			
Total	52	51	50
Interstitial	18	16	17
Terminal	34	35	33
Terminalization coefficient	0.654	0.686	0.660
1a Nos. of chiasmata per bivalent			
Total	1.27	1.19	1.14
Interstitial	0.44	0.37	0.39
Terminal	0.83	0.81	0.75
2 No. of bivalents with			
1-chiasma	30	35	38
per cent	73.2	81.4	86.4
2-chiasmata	11	8	6
per cent	26.8	18.6	13.6
3-chiasmata
per cent
3 Terminalization coefficient with			
1-chiasma	0.633	0.686	0.532
2-chiasmata	0.682	0.688	1.000
3-chiasmata

MORPHOLOGY OF THE SOMATIC CHROMOSOMES OF THREE ASIATIC COTTONS

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(Received for publication on 21 June 1939)

(With three text-figures)

THE morphology of the somatic chromosomes of the three Asiatic cottons, *G. Stocksii* Mast., *G. arboreum* L. var. *neglectum* forma *indica* H. & G. (strain K1) and *G. herbaceum* L. var. *frutescens* Delile (strain 2919) was studied in detail. Root-tips of germinating seeds were fixed in a number of fixatives of which Navashin's fluid was found most satisfactory. The slides were stained in Newton's Gentian violet and Haidenhain's haematoxylin. Both stains gave good results with the above fixing fluid. The lengths of the chromosomes in five metaphase plates have been measured in each species, from root-tips which were fixed the same day and have been given the same treatment. Care was taken to see that the metaphase plates selected for measurement were from approximately analogous portions of the roots and that in all these plates the chromosomes were well spread out with as few bends and curves in the individual chromosomes as possible. The lengths of chromosomes were measured with the aid of an eye-piece micrometer which was adjusted to small lengths so as to enable the bends in the chromosomes to be followed easily. The lengths were then converted into microns. Correction for for-shortening was not done because, as mentioned above, care was taken to see that the metaphase plates selected did not manifest much for-shortening of chromosomes. Table I gives the chromosome size frequencies in a single metaphase plate of each of the three species. For the sake of convenience chromosomes showing approximately the same length in each species have been grouped together in this table.

TABLE I
Chromosomes size frequencies of the three species

Species	Length variations in microns						Total No. of chromo- somes
	2.2	2.5	2.8	3.0	3.3	3.6	
<i>G. Stocksii</i>	6	10	6	4	26
<i>G. arboreum</i> , K1	6	10	8	2	26
<i>G. herbaceum</i> , 2919	2	8	6	8	2	26

The differences between the chromosome lengths of the three species have been statistically analysed and the results are given in Table II.

TABLE II

Analysis of variance of the chromosome lengths of the three species

Name of the species	No. of plates examined	Average length per plate	Error per cent	Whether z test was satisfied	Critical difference ($P=0.05$)
<i>G. herbaceum</i> . . .	5	78.3
<i>G. Stocksii</i> . . .	5	68.4	0.76	Yes	1.77
<i>G. arboreum</i> . . .	5	68.0

Conclusion :—*G. herbaceum* > *G. stocksii* = *G. arboreum*

The following conclusions regarding chromosome morphology have been recorded :

CHROMOSOME SIZE

(a) The three species studied showed a gradation in the lengths of the chromosomes (figs. 1a-3b) from the shortest to the longest as was also observed by Skovsted [1934] in *G. arboreum* L. (1.9μ — 3.2μ) and Arutjunova [1936] in *G. herbaceum* L. and *G. hirsutum* L. Baranov [1930] also observed differences in size of the respective arms of the chromosomes and of the satellites.

(b) The two species *G. arboreum* L. and *G. Stocksii* Mast. have been found to have the same range in the lengths of their chromosomes, which is about 2.2 microns to 3 microns whereas in *G. herbaceum* L., the range is from 2.5 to 3.6 microns, thus revealing a distinct increase in the lengths of all the chromosomes of *G. herbaceum* L. (Table I). The variations in the total length of chromosomes between the three species may be noted from Table II.

(c) There is distinct variation in the thickness of the chromosomes among the three species, although under identical fixation and treatment, the chromosomes of each species have been found to show uniform thickness (Figs. 1a—3b). The chromosomes of *G. Stocksii* Mast. are the thinnest, while those of *G. herbaceum* L. are the thickest, *G. arboreum* L. occupying an intermediate position. The differences in thickness indicate differences in volume. These variations in size from species to species may be considered to be genetic characters of the species.

1a



Somatic metaphase plate of *G. stocksii*
Mast. (x 5000)

2a

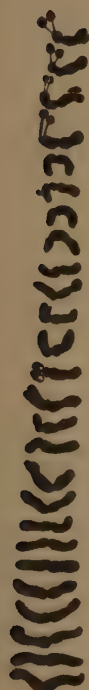


Somatic metaphase plate of *G. arboreum* L. var.
neglectum, forma *indica* (x 5000)

3a



Somatic metaphase plate of *G. herbaceum*
L. var. *frutescens* (x 5000)



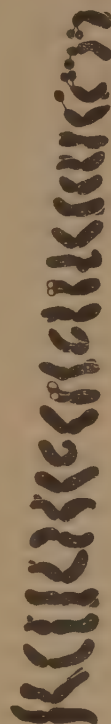
1b

Somatic chromosomes of *G. stocksii* Mast. arranged in a
line (x 5000)



2b

Somatic chromosomes of *G. arboreum* L. var. *neglectum*,
forma *indica*, arranged in a line (x 5000)



3b

Somatic chromosomes of *G. herbaceum* L. var. *frutescens*
arranged in a line (x 5000)

CHROMOSOME MORPHOLOGY

1. In all of the three species examined, two pairs of chromosomes were found to have satellites as was observed by Skovsted [1933, 1935] in these three species and Arutjunova [1936] in *G. herbaceum* L.

2. Attachment constrictions—

(a) The two pairs of chromosomes with satellites in all the three species appear to be morphologically distinct in that one pair has median attachment constriction whereas the other pair has sub-median attachment constriction at the satellite end.

(b) Of the rest of the chromosomes in each of the three species, two pairs have their attachment constrictions situated nearly a third of the length from one end. In *G. Stocksii* Mast. and *G. abroreum* L., these two pairs are among the medium-sized ones whereas in *G. herbaceum* L. one of these two pairs is found to be one of the longest (Figs. 1a—3b).

(c) All the other chromosomes have more or less median attachment constrictions.

According to Skovsted [1934], half of the chromosomes of the American cottons ($2n=52$) are small and the other half longer, the latter being comparable in size to the chromosomes of Asiatic cottons ($2n=26$), and the former to those of American wild species ($2n=26$). This led him to the conclusion that the American cottons ($2n=52$) originated from a cross between an Asiatic cotton and an American wild cotton ($2n=26$). He further states [Skovsted, 1935] that no difference in size and other features could be observed between the somatic chromosomes of the Asiatic cottons, *G. Stocksii* Mast., *G. abroreum* L. and *G. herbaceum* L. In the present investigation I have observed distinct differences in the size and other morphological features. These studies indicate the need for a re-examination of the problem relating to chromosome size and morphology in the different species of cotton and their hybrids.

ACKNOWLEDGEMENTS

This investigation was part of the work done by me as a research scholar of the Indian Central Cotton Committee under the guidance of Dr E. K. Janaki Amm M.A., D.Sc., Genetticist, Imperial Sugarcane Breeding Station, Coimbatore.

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ON THE NATURE OF THE REACTIONS RESPONSIBLE FOR SOIL ACIDITY

VI. THE VARIABILITY OF THE TOTAL NEUTRALIZABLE ACID OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS*

BY

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(With seven text-figures)

IN part V of this series [Mitra, 1936] experimental results have been given showing the indefinite character of the total neutralizable acid of a hydrogen clay sol calculated from its titration curves. Using different bases, the titration curves gave different values of the total acid measured at a fixed pH . In this paper, the variations of the total acid have been studied in greater detail. The total acid has been estimated on titration in presence and absence of neutral salts. Different concentrations of salts have been used and the titrations carried out with different bases. The total acids of the corresponding ultrafiltrates and leachates have also been measured.

Such investigations have a twofold interest. They are likely, first, to elucidate the electrochemical behaviour of hydrogen clay sols as colloidal acid systems and secondly, to bring out the factors affecting the total amount of acid which enters into the reaction between a soil and an added base, that is, the factors which affect the base binding capacity and the lime requirement of soil. These quantities are rather illdefined [Hissink, 1935], concordant results being seldom obtained by alternative routine methods [Crowther and Martin, 1925]. A knowledge of the factors responsible for the variations is thus necessary. One of the objects of this investigation has been the elucidation of these factors.

Experimental

The method of preparing colloidal solutions of hydrogen clays, the technique of potentiometric and conductometric titrations and the various apparatus used in this work have been described in the previous paper of this series

*The results given in this paper have been taken from the published annual reports for 1935-36 and 1936-37 on the working of a 'Scheme of Research into the Properties of Colloid Soil Constituents' financed by the Imperial Council of Agricultural Research, India and directed by Professor J. N. Mukherjee. The authors' thanks are due to the University of Calcutta for permission to work in the Physical Chemistry Laboratories of the University and for other facilities.

**Senior Assistant Soil Chemist under the above scheme.

[Mitra, 1936]. The soils* from which the hydrogen clays used in this work were prepared are listed below together with the available information regarding them.

1. Suri (Bengal) Farm Soil. Agricultural Chemist's experimental plot. Block A 1—16, plot Nos. 3, 5, 16. No manure. Collected from a depth of 6-in.—12-in.
2. Burdwan (Bengal) Farm Soil. Block B, plot No. 40. Standing crop—Kalai; surface soil collected from a depth of 0-in.—6-in.
3. Soil from Government Seed Farm, Kalyanpore (U. P.); a brown loam; surface soil included in the 'Doab'.
4. Black Cotton Soil from Satara Dt. (Bombay Province); surface soil, calcium saturated and neutral.
5. Black Soil from Bilaspur near Raipur (Central Provinces); surface soil, neutral.
6. Red laterite soil from Dacca (Bengal) Farm collected from a depth of 0-in.—6-in.

Aqueous suspensions of hydrogen clays prepared from the clay fractions of the first three soils were centrifuged to obtain stable sols, E, F and H respectively. Sols I, K and L were prepared from hydrogen clays obtained, respectively, from the entire clay fractions of the remaining three soils.

Results

A. TOTAL ACIDITIES OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS OBTAINED ON TITRATION WITH DIFFERENT STRONG BASES

Figs. 1, 2 and 3 show the titration curves of sols E, H and L obtained on titration with different bases.

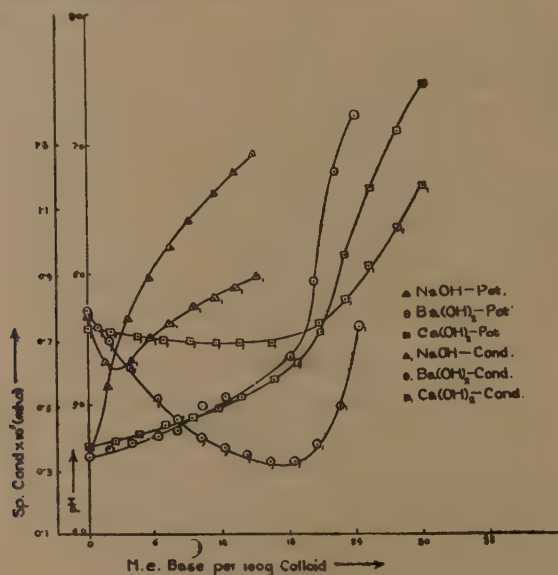


FIG. 1. Sol E

*Soils Nos. 1, 2 and 6 were obtained through the courtesy of the Agricultural Chemist to the Government of Bengal. Soils Nos. 3, 4 and 5 were kindly supplied by the Superintendent, Government Seed Farm, Cawnpore and the Agricultural Chemists to the Governments of Bombay and C. P. respectively.

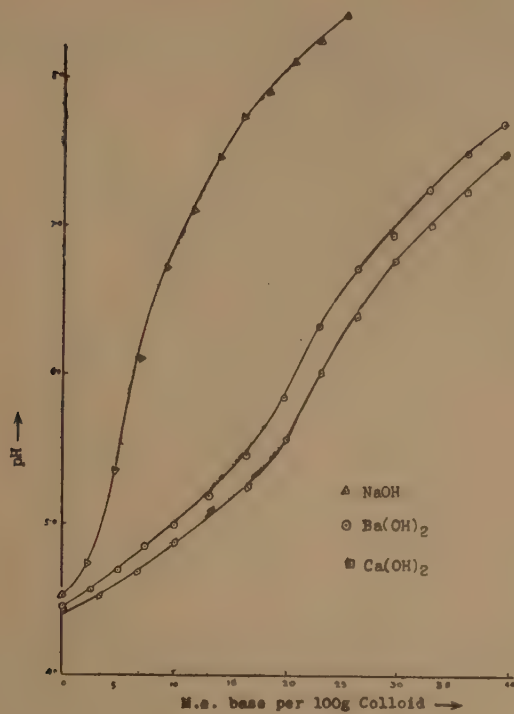


FIG. 2. Sol H

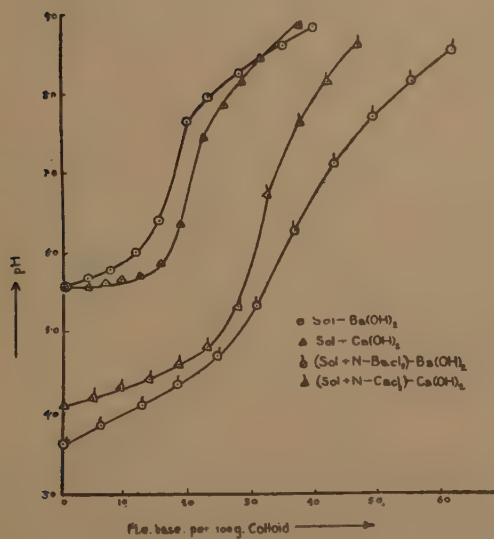


FIG. 3. Sol L

In agreement with previous observations [Mitra, 1936] the titration curves* show definite inflection points and minima. The total acids of the sols calculated from the titration curves at the first ** inflection point and the minimum as also at pH 7.0 are given below.

TABLE I
Total acid in m.e. base per 100 gm. colloid (oven-dried)

Sol	Base used for titration	At first inflection point of titration curve	At pH 7.0	At minimum of conductometric curve
E	Ba (OH) ₂	20.6	25.0	19.6
	Ca (OH) ₂	21.5	26.2	18.0
	NaOH	2.2	15.4	2.5
H	Ba (OH) ₂	21.5	32.0	n.d.*
	Ca (OH) ₂	21.5	32.8	n.d.
	NaOH	10.7	n. d.
L	Ba (OH) ₂	17.5	17.0	n. d.
	Ca (OH) ₂	19.0	19.5	n. d.

* Not determined.

The total acidity of soils E and H at the inflection point, or, the minimum of the conductometric titration curve is less than that at pH 7.0. This is to be expected as the inflection points and the minima occur in the acid region.

The total acidities at the inflection point and at pH 7.0 which react with different bases are in the order $\text{Ca(OH)}_2 > \text{Ba(OH)}_2 > \text{NaOH}$. Calcium hydroxide appears to have a somewhat greater effect than baryta. The relative effects of these two bases have been more fully dealt with later.

B. EFFECT OF ADDITION OF NEUTRAL SALTS ON THE TOTAL ACIDITY OF HYDROGEN CLAY SOLS

The nature of the titration curve of a hydrogen clay sol with a given base as also the total acidity calculated from the curve have been observed in this work to be modified to a marked extent when the titration is carried out in the presence of a neutral salt. This 'neutral salt effect' has been studied in some detail. For this purpose, the total acidities of (i) the sols, (ii) the sol + salt mixtures, (iii) the clear supernatant liquids above the coagula of these mixtures and (iv) the neutral salt extracts obtained on repeatedly leaching the sols with solutions of the salts have been estimated. Different bases and different neutral salts have been used.

*A detailed discussion of the features of the curves has not been entered into in this paper. This will be done in the concluding paper of this series to be shortly communicated for publication in this journal.

**The titration curves do not show a second inflection up to the pH to which the titration had been extended. It might be observed at higher alkaline regions where, however, a 'break-down' of the exchange complex might occur. Evidence of a complete 'break-down' of the H-clay from a lateritic soil has been obtained at pH 13.5 though no 2nd inflection was observed on titration to this pH. Further work on this point is in progress.

(a) *Total acids in presence and absence of salts having the same cations as the bases*

The following results were obtained with sols E, F, H and L.

TABLE II

Sol	Base used for titration	Ta*	Ta'*	Salt added and conc. of salt	Ts*	Ts'*	Ts/Ta	Ts'/Ta'	** pH(a)	** pH(s)
E	Ba(OH) ₂	20.6	25.0	0.1N BaCl ₂	28.0	>42.4	1.31	>1.67	6.0	4.6
	Ca(OH) ₂	21.5	26.2	0.1N CaCl ₂	21.2	40.6	0.97	1.57	5.8	4.1
	NaOH	2.2	15.4	0.1N NaCl	16.1	26.4	7.30	1.71	5.4	5.0
F	Ba(OH) ₂	31.0	39.4	0.1N BaCl ₂	38.4	64.1	1.24	1.63	6.2	5.2
	NaOH	9.8	32.0	0.1N NaCl	27.0	56.0	2.70	1.75	5.0	4.8
H	Ba(OH) ₂	21.5	32.0	0.83N BaCl ₂	30.5	50.0	1.42	1.56	5.8	4.8
	Ca(OH) ₂	21.5	32.8	0.83N CaCl ₂	29.3	47.0	1.86	1.43	6.6	5.0
	NaOH	...	10.7	0.83N NaCl	22.5	40.0	...	3.8	...	4.8
L	Ba(OH) ₂	17.50	17.0	1.0N BaCl ₂	32.0	40.5	1.82	2.31	7.1	5.4
	Ca(OH) ₂	19.0	19.5	1.0N CaCl ₂	30.0	32.5	1.58	1.66	6.6	6.1

*Ta and Ta' denote the total acidities in m. e. base per 100 gm. of colloid in absence of salt calculated respectively from the first inflection point of the potentiometric titration curve and from the amount of the base which had to be added to make the pH of the sol 7.0. Ts and Ts' denote the corresponding total acidities in presence of salt.

**pH(s) and pH(a) denote the pH at the first inflection point in the titration curve obtained on titration in presence and absence of salt respectively.

Figs. 3, 4, 5 and 6 show the titration curves from which the total acidities given in table II have been calculated.

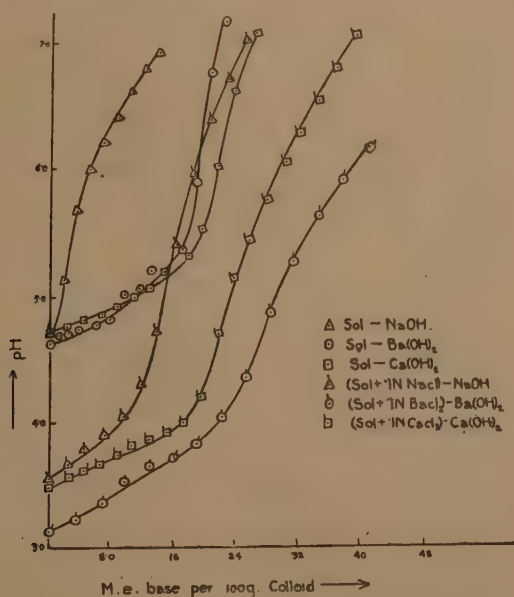


Fig. 4. Sol E

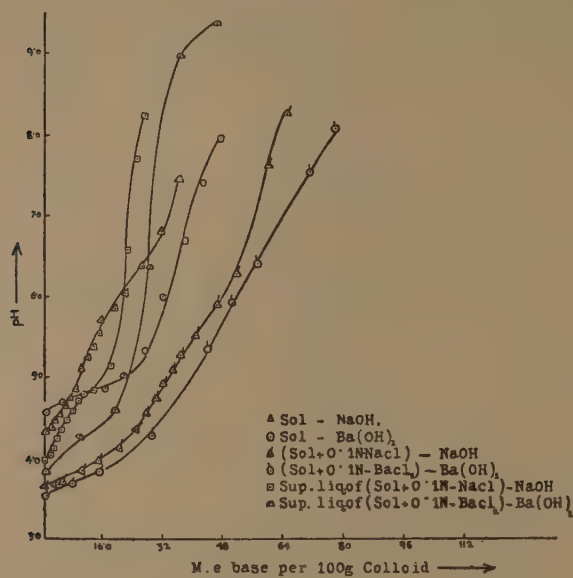


FIG. 5. Sol F

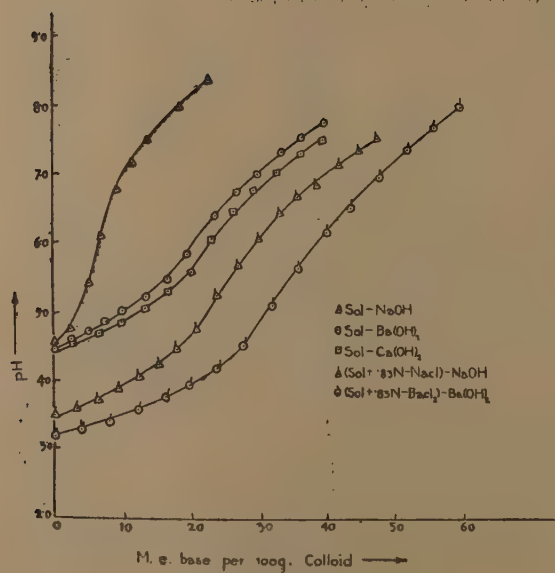


FIG. 6. Sol H

The results show that a considerably larger value of the total acid calculated at the inflection point of the titration curve is obtained when the sol is titrated in the presence of a salt than when titrated alone. In making this comparison, the pH at which the total acids are measured is an important factor for, as the titration curves show, increasing amounts of the acid react with the base as the pH rises. The inflection point in the titration curve of a sol + salt mixture occurs at a lower pH than the inflection point in the titration curve of the sol itself (Table II). It is significant that even then a larger value of the total acid (calculated at the inflection point) is obtained on titration in the presence of the salt than in its absence. The cations of the salt present in large numbers thus have an unmistakable effect on the total amount of the acid reacting with the base. This *cation effect* is as important a factor in determining the total acid as the pH at which it is measured. It is strikingly brought out on comparing the total acids of the sol (Ta') and the sol + salt mixture (Ts') at the same pH , e.g. pH 7.0. Ts' is always greater than Ta' .

Table II shows a slightly lower value of the total acid (at the inflection point) of sol E when it is titrated with $Ca(OH)_2$ in presence of $0.1 N$ $CaCl_2$ than when titrated alone. The difference is only ± 1.5 per cent and lies within the limits of experimental error. The failure to obtain a larger total acid in the presence of the salt arises mainly from the fact that the inflection point in the titration curve occurs at a much lower pH when the sol is titrated with the salt than when the sol alone is used. Measured at pH 7.0, the sol + $CaCl_2$ mixture shows a much larger total acid than the sol itself indicating a distinct cation effect.

Titration with caustic soda in the presence of $NaCl$ shows the highest relative increase (highest value for Ts/Ta) in the total acid calculated at the first inflection point. This signifies that at the inflexion point caustic soda neutralizes only a small fraction of the H^+ ions associated with the colloidal particles. It neutralizes them after they have been exchanged for the cations of the salt.

A reference to Table II will further show that the total acid (both at the first inflection point and at pH 7.0) reacting with different bases in the presence of the corresponding salts at the same concentration follows the order: $Ba(OH)_2 > Ca(OH)_2 > NaOH$. The order of total acidities, however, changes to $Ca(OH)_2 > Ba(OH)_2 > NaOH$ when the sols alone are titrated.

The following results illustrate the dependence of the total acid on the concentration of the added salt.

TABLE III

Sol	Base used for titration	Salt added	Conc. of salt	Ta^*	Ta'^*	Ts^*	Ts'^*	Ts/Ta	Ts'/Ta'
E	NaOH	NaCl . .	0.01N .	2.2	15.4	16.4	19.5	7.45	1.27
	NaOH	NaCl . .	0.1N .	2.2	15.4	16.1	26.4	7.30	1.71
	$Ba(OH)_2$	$BaCl_2$. .	0.01N .	20.6	25.0	20.8	25.5	1.00	1.02
	$Ba(OH)_2$	$BaCl_2$. .	0.1N .	20.6	25.0	28.0	>42.4	1.31	1.67
H	$Ba(OH)_2$	$BaCl_2$. .	0.05N .	21.5	32.0	25.0	38.0	1.16	1.19
	$Ba(OH)_2$	$BaCl_2$. .	0.25N .	21.5	32.0	27.0	44.0	1.26	1.37
	$Ba(OH)_2$	$BaCl_2$. .	0.83N .	21.5	32.0	30.5	50.0	1.42	1.56

* Ta , Ta' , Ts and Ts' have the same significance as indicated before.

Fig. 7 shows the titration curves of sol H and sol H + BaCl₂ mixtures from which their total acids given in table III have been calculated.

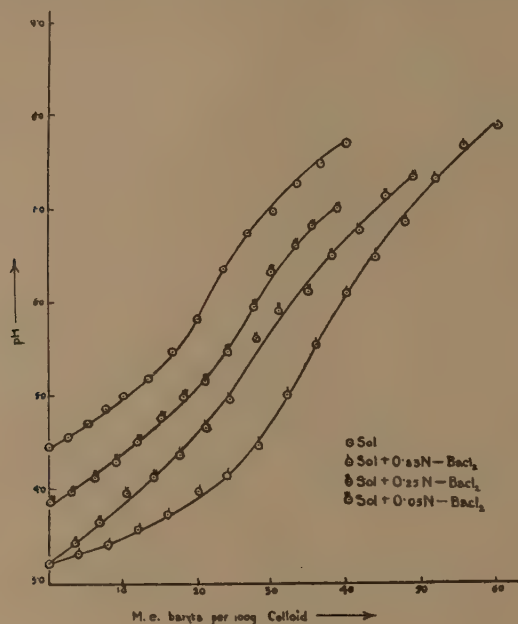


FIG. 7, Sol H

Beginning with the titration curve of the sol at the extreme left a lateral displacement of the curves of the sol + salt mixtures towards the right is observed as the concentration of the salt in the mixture increases indicating a gradual increase in the total acidity with the concentration of the salt in the mixture.

(b) *The total acidities of (i) the sol, (ii) the sol + salt mixture and (iii) the clear supernatant liquid above the coagulum of the sol + salt mixture*

In Fig. 5 are given the titration curves of (i) sol F, (ii) mixtures of sol F and salts (0.1N) and (iii) the clear supernatant liquids above the coagula of these mixtures.

The inflection points in the titration curves of (iii) are sharper than those of (i) and (ii) which signifies that a strong acid is being neutralized in (iii). The titration curves of (iii), however, show a slight initial rise followed by a small region of stronger buffering before the sharp inflection is observed. The initial rise would not be expected if only hydrochloric acid formed by the interaction of the hydrogen clay and the neutral salt were being titrated in (iii). It would be expected if (iii) also contained some 'displaced' aluminium ions. In the next paper of this series it has been shown that using a sufficiently

low concentration of the salt, preferably that of an alkali metal cation, the titration curve of (iii) has a truly strong acid character, the above initial rise not being in evidence. Also, actual analysis has shown that under these conditions (iii) contains practically no Al^{+++} ions.*

The total acidities calculated from the titration curves given in fig. 5 are shown in the following table.

TABLE IV

System	Total acid in m. e. Ba (OH) ₂ per 100 gm. of colloid	
	At first inflection point	At pH 7.0
(i) Sol F	31.0	39.4
(ii) Sol F + BaCl ₂ mixture	38.4	64.1
(iii) Supernatant liquid of (ii)	30.9

The total acidities are in the order (ii) > (i) > (iii). The difference between the total acids of (ii) and (iii) signifies that at the concentration used, the salt does not displace all the hydrogen ions associated with the colloidal particles into the intermicellary liquid of the sol and that a part of the hydrogen ions brought into a reactive condition by the neutral salt still remains associated with the colloidal particles and these are capable of reacting with the base.

(c) *The total acidity of (i) the sol, (ii) the sol + salt mixture and (iii) the neutral salt extract obtained by repeatedly leaching the sol with the salt solution.*

Leaching with neutral salts is often resorted to in the estimation of the lime requirement of soil by the titration of the salt extract [Hopkins, 1903; Daikuhara, 1914; Gedroiz, 1924]. It has been shown above that the addition of the salt to the sol does not displace into the intermicellary liquid all the H^+ ions which are brought into a reactive, that is, neutralizable condition and which can, therefore, be estimated only by titrating the sol + salt mixture *in situ*. It was thus of interest to compare the amount of acid displaced by repeated leaching of the sol with the salt solution. This has been done with sol H. The solution with which the sol was leached had the same concentration (0.83N) as obtained in the sol + salt mixture. The following results were obtained.

*The subject is being systematically studied by Mr. B. Chatterjee in this laboratory.

TABLE V

System	Total acid in m. e. base per 100 gm. colloid using			
	Ba(OH) ₂ and BaCl ₂		NaOH and NaCl	
	At first infection pt.	At pH 7.0	At first infection pt.	At pH 7.0
Sol	21.5	32.0	..	10.7
Sol + Salt	30.5	50.0	22.5	40.0
1st 100 c.c. of leachate	17.0	..	15.0
2nd 100 c. c. of leachate	3.0	..	2.0
3rd 100 c.c. of leachate	1.2	..	1.8

In agreement with the results given in Table II the total acidity of the sol + salt mixture is greater than that of the sol alone. The sum of the total acidities of the three leachates, however, is less than the total acidity of the sol + salt mixture. Of the three leachates, the total acidity of the first is the highest, then there is a sudden drop, the total acid of the third leachate being almost negligible. Thus treatment of the hydrogen clay sol with the salt solution, to the extent that practically no further acid comes out in the leachate, does not displace from the colloidal particles of the sol all the acid which can react with the base in presence of the salt. The salt and the base when they can react together liberate the greatest amount of acid under the conditions compared. *Of this amount a part is liberated into the intermicellary liquid and another part remains associated with the colloidal particles.*

Table V shows that when leached with barium chloride a greater amount of colloid-free acid is obtained in the extract than with sodium chloride using equal concentrations. This is in agreement with the greater total acidity (previously observed) of the sol + salt mixture obtained on titration with Ba(OH)₂ than NaOH in presence of BaCl₂ and NaCl respectively.

C. THE RELATIVE EFFECTS OF Ba⁺⁺ AND Ca⁺⁺ IONS IN THE INTERACTIONS OF THEIR SALTS AND BASES WITH HYDROGEN CLAYS

The results given in the preceding sections show that in the interactions of hydrogen clays with bases both in the presence and absence of neutral salts a definite cation effect exists. In the interactions in presence of salts the relative effects of the cations agree with the lyotrope series and Ba⁺⁺ has an unmistakably greater effect than Ca⁺⁺. Previous work from this laboratory [Mitra, 1936] shows that the H⁺ ion activity of a hydrogen clay sol shows characteristic variations on progressive additions of a neutral salt and the

relative effects of different salts having a common anion also follow the lyotrope series, that is, Ba^{++} has a greater effect than Ca^{++} . The following results further illustrate this point.

TABLE VI

Sol	Conc. of the added salt $\times 10^3N$	Lowering of pH	
		With BaCl_2	With CaCl_2
H	1.5	0.48	0.45
	3.0	0.51	0.49
	40.5	0.87	.77
I	1.0	0.67	0.65
	5.0	.82	.73
	45.0	1.09	1.03
I'	1.5	0.65	0.60
	6.8	.84	.81
	50.5	1.14	1.02
K	1.20	0.65	0.61
	9.00	.95	.90
	42.00	1.19	1.12
K'	1.20	0.68	0.61
	9.00	.88	.81
	42.00	1.12	1.00

Hydrogen clays I' and K' were obtained from the same soils as hydrogen clays I and K with the difference that the colloidal materials of these soils were treated according to the method of Drosdoff and Truog [1935] to remove their free silica, alumina and ferric oxide before converting them into hydrogen clays I' and K'.

Table VI shows that barium chloride lowers the pH of the sol to a greater extent than calcium chloride at the same concentration. The difference in the relative effects of the two ions persists even after removal of free silica and sesquioxides from the hydrogen clay as the results obtained with sols I' and K' show.

Reference has already been made to the fact that calcium hydroxide reacts more strongly than baryta with a hydrogen clay sol to which no salt has been added indicating a greater relative effect of Ca^{++} than Ba^{++} ions under these conditions. This is further illustrated by the following results.

TABLE VII

Sol	Base used for titration	pH at inflection point	Total acid in m. e. base per 100 gm. of colloid		
			At inflection point.	At pH 7.0	At pH 9.0
H	$\text{Ba}(\text{OH})_2$	5.80	21.5	32.0	..
	$\text{Ca}(\text{OH})_2$	6.60	21.5	32.8	..
I	$\text{Ba}(\text{OH})_2$	7.00	82.0	82.0	101.5
	$\text{Ca}(\text{OH})_2$	6.95	96.0	97.0	122.0
I'	$\text{Ba}(\text{OH})_2$	7.60	91.0	85.0	106.0
	$\text{Ca}(\text{OH})_2$	6.50	86.0	91.0	114.0
K	$\text{Ba}(\text{OH})_2$	5.80	55.0	61.0	81.0
	$\text{Ca}(\text{OH})_2$	5.20	58.0	67.0	86.0
K'	$\text{Ba}(\text{OH})_2$	5.75	56.0	62.0	75.5
	$\text{Ca}(\text{OH})_2$	5.78	63.0	68.5	81.5

Calcium hydroxide reacts with the sol more strongly than baryta even beyond the neutral point (at pH 9.0). This difference in the relative effects of Ba^{++} and Ca^{++} ions in the interactions of their salts and bases with hydrogen clays illustrates *two different types of cation effect* discussed in the next section.

D. THE ROLE OF THE ELECTRICAL DOUBLE LAYER AND OF THE SECONDARY ADSORPTION OF CATIONS IN DETERMINING THE TOTAL NEUTRALIZABLE ACID OF A HYDROGEN CLAY SOL

The results previously given bring out the variable character of the total neutralizable acid of a hydrogen clay sol. The total acid is a function (1) of the pH at which it is measured and (2) of 'cation effects'. The cation effect finds expression in the different total acids, measured at the same pH, obtained on titration with different bases as also in the considerably larger total acid obtained on titrating the sol in the presence of a neutral salt than on titrating it alone. Such variations in the total acid are not possible for any truly dissolved acid.

The theory of the electrical double layer postulating the existence of primarily adsorbed ions associated with the colloidal particles of the sol and of a secondary adsorption of cations by them [Mukherjee, 1921, 1922] affords a satisfactory basis for an interpretation of the cation effects and the variations

of the total acid.* According to the theory, H^+ ions corresponding to the primarily adsorbed anions which are assumed to be 'built in' on the solid side of the solid-liquid interface may exist in two states, viz. (1) in a secondarily adsorbed condition either by simple electrostatic forces, or, by specific forces of the chemical or forces of the Van der Waals' type and (2) in a free, or, 'mobile' state which determines the free charge of the surface. The distribution between the two states depends on the nature of the colloid and that of the intermicellary liquid. The H^+ ions of both categories may be displaced by the cations of an added salt or a base, these cations being themselves adsorbed in the process; the displacement of H^+ ions of the second category is obviously the easier. The adsorption, i.e. fixation of the cations leads to the formation of 'ion pairs' on the surface and is brought about by electrostatic forces of attraction and/or by specific forces, e.g. of a chemical or Van der Waal's type. In the former case, only the electrical properties of the cations, e.g. their valency, mobility and state of hydration (which determines the distance between the centres of the ions constituting the ion pairs formed by adsorption) are the factors which determine the intensity of their adsorption and hence the amount of H^+ ions exchanged. In the interaction with a base, these exchanged H^+ ions are neutralized by the OH^- ions of the latter. The possibility of a direct neutralization of some H^+ ions by the OH^- ions is not excluded. The greater total acidity measured at a given pH obtained on titration with baryta or calcium hydroxide compared to caustic soda is, on this view, due to a greater electrical adsorption of Ba^{++} and Ca^{++} ions compared to that of Na^+ ions. A greater total acid obtained on titration with a given base in the presence of a salt than in its absence similarly arises from a stronger adsorption of the cations because of their being present in larger numbers.

The increase in total acid observed on titration in the presence of a salt may be due to aluminium ions which, as our later work shows, are always present in the supernatant liquid above the coagulum of the sol + salt mixture whenever such an increase in the total acid is observed. The aluminium ions may have been directly exchanged for the cations of the salt, or, they may have been dissolved out from the hydrogen clay by the free acid liberated by its interaction with the salt. The mechanism of the liberation of such aluminium ions is of no direct significance so long as discussions of the cation effects are restricted, as has been done here, to their role in determining the total neutralizable acids of the sol + salt mixtures and their clear supernatant liquids.

The difference in the relative effects of Ba^{++} and Ca^{++} ions in the interactions of their salts and bases with hydrogen clays may be explained as follows. The greater relative effect of Ba^{++} compared to Ca^{++} in the interactions with the salts arises from a stronger electrical adsorption of Ba^{++} than Ca^{++} , the relative electrical adsorbability of the ions being determined by their electrical properties, viz. their valency, mobility and state of hydration. The cation effect is thus electrical in origin and follows the

*The picture here suggested is of a general nature. It takes no account of (i) the amphoteric character of the hydrogen clays (ii) the role of Al^{+++} and other ions on the surface in addition to H^+ ions and (iii) the detailed structure of the hydrogen clays. Work covering these aspects is under way.

lyotrope series. It may thus be called a *regular cation effect*. The greater relative effect of Ca^{++} than Ba^{++} ions in the interactions with the bases is an '*irregular or specific cation effect*' in the sense that it does not follow the lyotrope series. This second type of cation effect' does not result from simple electrical adsorption (by electrostatic forces) of the cations together with their hydration envelopes. Here, the cations are adsorbed by specific forces other than simple electrostatic forces in a dehydrated state. The two types of cation effect will be further discussed in the concluding paper of this series.

Summary

The total neutralizable acid of a hydrogen clay sol calculated at the first inflection point and the minimum of its potentiometric and conductometric titration curves with bases as also at $p\text{H}$ 7.0 is a variable quantity. The variations arise from cation effects whose nature has been discussed in the light of the theory of the electrical double layer and of adsorption of ions.

On titration with different bases in absence of salts, the total acid decreases in the order $\text{Ca}(\text{OH})_2 > \text{Ba}(\text{OH})_2 > \text{NaOH}$. The greater relative effect of $\text{Ca}(\text{OH})_2$ compared to $\text{Ba}(\text{OH})_2$ illustrates an '*irregular, or, specific cation effect*'

With a given base, titration in the presence of a salt yields a much larger total acid than titration of the sol alone. The cations present in large numbers have an effect.

On titration in presence of the same concentration of the corresponding chloride, the total acid decreases in the order $\text{Ba}(\text{OH})_2 > \text{Ca}(\text{OH})_2 > \text{NaOH}$. Here, a '*regular cation effect*' is observed which is in agreement with the (lyotrope) series.

The total acid of the sol+salt mixture obtained on titrating it *in situ* is greater than that of (i) the clear supernatant liquid above the coagulum of the mixture as also (ii) the salt extract obtained on repeatedly leaching the hydrogen clay with the solution of the salt. A considerable amount of titratable acid thus remains associated with the coagulum of the mixture which cannot be displaced in the intermicellary liquid of the sol even on repeatedly leaching it with the solution of the salt.

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VII. THE ELECTROCHEMICAL PROPERTIES OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS*

BY

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(With ten text-figures)

IN Part V of this series [Mitra, 1936] mention has been made of several points of difference which exist between the titration curves of colloidal solutions of hydrogen clays and those of acids in true solution. In part VI [Mitra, Mukherjee and Bagchi, 1940] it has been shown that the total acidity of a hydrogen clay sol calculated from titration curves obtained on titrating the sol under different conditions, e.g. with different bases or, with a given base but in the presence and absence of a neutral salt, is, also unlike any dissolved acid, a variable quantity. In this paper, the special features of hydrogen clay sols which distinguish them from acids in true solution have been considered in greater detail and an attempt has been made to reconcile them in a general picture which is based on the theory of the electrical double layer and of adsorption of ions.

Experimental

The hydrogen clay sols used were prepared from the following Indian soils† in the manner previously described [Mitra, 1936].

Sol	Soil from which the hydrogen clay was obtained
D	Virgin soil from Dacca Farm (Bengal) collected from a depth of 0 to 6 in.
E	Suri (Bengal) farm soil. Agricultural Chemist's experimental plot. Block A 1-16, plot Nos. 3, 5, 16. No manure; soil collected from a depth of 6 to 12 in.
F	Burdwan (Bengal) Farm soil. Block B, plot No. 40. Standing crop— <i>kalai</i> ; surface soil collected from a depth of 0 to 6 in.
G	Krishnagar (Bengal) farm soil; highland soil collected from a depth of 0 to 6 in.
H	Soil from Government Seed Farm, Kalyanpore (U. P.); a brown loam included in the <i>Doab</i> ; collected from a depth of 0 to 6 in.
I	Black cotton soil from Satara Dt., Bombay Presidency; surface soil, calcium saturated, neutral.
K	Black soil from Bilaspur near Raipur (C. P.); surface soil.

* The results given in this paper have been taken from the published annual reports for 1935-36 and 1936-37 on the working of a scheme of 'Research into the Properties of Colloid Soil Constituents' financed by the Imperial Council of Agricultural Research, India, and directed by Professor J. N. Mukherjee. The author takes this opportunity to thank the University of Calcutta for permission to work in the Physical Chemistry laboratory of the University and for other facilities.

** Senior Assistant Soil Chemist under the above scheme.

† The first four soils were kindly supplied by the Agricultural Chemist to the Government of Bengal. The Kalyanpur Farm Soil was obtained through the courtesy of the Superintendent, Government Seed Farm, Cawnpore. The two black soils were kindly supplied by the agricultural chemists to the Governments of Bombay and the Central Provinces, respectively.

The experimental arrangements and procedure were as previously described [Mukherjee *et. al.*, 1936 ; Mitra, 1936].

The hydrogen clay sols used covered a range of H ion concentrations from 10^{-5} *N* to 10^{-4} *N* and had specific conductivities of the order of 10^{-5} to 10^{-6} mho. The significance and degree of accuracy of estimations of free and total acids of such systems have been discussed in two previous parts of this series [Mukherjee, *et. al.*, 1936 ; Mitra, 1936]. Since the publication of these papers, it has been possible to further improve upon the reproducibility and accuracy of the *pH* data [Mukherjee, Mitra and Mukherjee, 1937] by more careful attention to preparation and cleansing of the electrodes and the inclusion of glass electrodes* used in conjunction with a valve potentiometer.** The results given in Table I obtained with some hydrochloric acid solutions which had nearly the same *pH* as the sols illustrate the order of accuracy now attained. The first, second and third columns of the table give the *pH* obtained with the hydrogen (*h*), glass (*g*) and quinhydrone (*q*) electrodes respectively. The fourth column gives the *pH* calculated from the observed specific conductivity from the known mobilities of H^+ and Cl^- ions and assuming complete dissociation of the acid. The fifth column gives the value of $\log \frac{1}{[Cl^-]}$ the Cl^- ion concentration having been determined by conductometric titration with silver sulphate solution. The sixth column gives the *pH* corresponding to the total acidity calculated from the potentiometric or conductometric titration curve of the acid assuming complete dissociation. Hydrogen and glass electrodes were used for the potentiometric titrations.

TABLE I

	<i>pH</i> (h)	<i>pH</i> (g)	<i>pH</i> (q)	<i>pH</i> (cond.)	Total Cl og $\frac{1}{[Cl]}$	<i>pH</i> (T)
Average :	2.94	2.94	2.96	3.08	2.96	3.00 (glass electrode)
	2.96					3.02 (hydrogen electrode)
	2.96					
Average :	4.34	4.30	4.30	4.42	4.41	4.40 (glass electrode)
	4.36					4.42 (hydrogen electrode)
	4.36					4.42 (conductometric)
	4.32					4.41
	4.35					
Average	5.12	5.08	5.13	5.23	Average	5.18 (glass electrode)
	5.10					5.14 (conductometric)
	5.11					
	5.11					5.16

* Morton type glass electrodes were used.

** A Cambridge *pH* meter reading directly to 2 millivolts.

The average *pH* values obtained with the hydrogen electrode agree with the value obtained with the glass or quinhydrone electrode within 1 per cent. The average of glass, quinhydrone and hydrogen electrode values for the three solutions are respectively 2.95, 4.32 and 5.11. These values agree with the respective total acidity values within 2 per cent.

With the experimental arrangement used, some KCl from the salt bridge has been occasionally found to find passage into the titration vessel thereby increasing the conductivity of the solution. Consequently, for accurate determinations of the absolute values of the conductivities and for studying the fine features of the conductometric titration curves, the conductometric measurements were carried out separately from the potentiometric measurements thus obviating the use of the KCl-bridge.

Results

I. HYDROGEN CLAY SOLS ARE NOT HOMOGENEOUS ACID SYSTEMS

In the following table the free and total acids of a number of hydrogen clay sols have been compared with those of their ultrafiltrates. The free acid has been calculated from the observed *pH* and the total acid from the inflexion point of the potentiometric titration curve obtained on titration with barium hydroxide.

TABLE II

System	<i>pH</i>	Free acidity (H ion conc. $\times 10^5$)	Total acidity (<i>N</i>) $\times 10^5$
Sol D	5.40	0.40	5.0
Ultrafiltrate of sol D	5.95	0.11	Negligible
Sol E	4.66	2.19	24.30
Ultrafiltrate of sol E	6.10	0.08	Negligible
Sol F	4.41	3.89	38.0
Ultrafiltrate of sol F	5.90	0.13	Negligible
Sol G	4.57	2.69	40.0
Ultrafiltrate of sol G	5.85	0.14	<i>n. d.*</i>
Sol H	4.52	3.02	99.0
Ultrafiltrate of sol H	6.05	0.09	<i>n. d.*</i>

* Not determined

The free and total acids of the ultrafiltrate are considerably less than those of the sol itself. A separation of the colloidal particles from the liquid phase by ultrafiltration thus causes a marked lowering of its H^+ ion activity and total acidity. A two phase acid character of the sol, the colloidal particles constituting one phase and the intermicellary liquid the other, is thus brought

out. There are osmotically active H^+ ions associated with the colloidal particles which, rather than any H^+ ions present in a truly dissolved condition in the intermicellary liquid, are responsible for the observed H^+ ion activity and total acidity of the sol. The observations are in agreement with the so-called 'suspension effect' of Wiegner and Pallmann [1929].

II. DISCREPANCIES BETWEEN CONDUCTIVITY AND ACTIVITY MEASUREMENTS WITH COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS

The peculiar phasal condition of the H^+ ions associated with the colloidal particles of a hydrogen clay sol is further illustrated by the following significant discrepancies between the specific conductivities of the sols actually measured and those calculated from their activity data. The results obtained with sols H_1 and J and with sols prepared by diluting them with their respective ultrafiltrates are given in Table III. The ultrafiltrates had pH values 6.09 and 6.20. Sol H_1 was obtained from the same soil as sol H. Only, it had a higher colloid content than sol H.

TABLE III*

Sol	H^+ ion conc. in normality $\times 10^5$	Sp. conductivity in mho $\times 10^5$ (Observed)	Sp. conductivity in mho $\times 10^5$ (Calculated)
H_1	8.7	1.02	3.48
H_2	5.01	0.97	2.04
H_3	3.16	0.61	1.26
J	14.45	2.20	5.16
J/4	3.72	0.88	1.37
J/6	2.57	0.66	1.17

The H^+ ion concentrations were calculated from the observed pH values of the sols measured with hydrogen and glass electrodes. For the conductivity measurements, a Washburn cell having a cell constant 0.0095 in conjunction with a Vreeland Oscillator as the source of the alternating current was used.

* The results given in this table have been taken from a paper read at a meeting of the Chemistry Section of the Indian Science Congress Association, held in January, 1938. An abstract of the paper has appeared in the Proceedings of the Congress (Proceedings, 1938, Vol. 3, p. 53).

The values of the calculated specific conductivity given in the above table were obtained with the aid of the equation $\mu_{\text{cal}} = C_{\text{H}^+} U_{\text{H}^+}$ where U_{H^+} is the equivalent conductance of H^+ ions and C_{H^+} , the H^+ ion concentration** calculated from the pH . The conductivity due to the anions, that is, the negatively charged colloidal particles has been neglected. The results given in Table III reveal the interesting fact that the actual specific conductivity is less than even the conductivity due to the H^+ ions calculated from their observed activities. These H^+ ions are not present in the intermicellary liquid of the sol as is shown by the fact that while the sols have H^+ activities of the order of $10^{-4}N$ to $10^{-5}N$ those of their ultra-filtrates are of the order of $10^{-7}N$. The results, therefore, show that though these H^+ ions register their activity on a reversible electrode, they do not take part in the conduction of electricity in the usual manner. Their average conductivity coefficient is much less than unity [Mukherjee, 1933]. This behaviour of hydrogen clay sols has not been previously observed* and it constitutes a peculiar feature of such systems.

Discrepancies between conductivity and activity measurements in colloidal solutions have been used by Mukherjee [1933] as an argument in favour of the existence of an electrical double layer in such systems. Hartley [1935] has sought to reconcile them in the light of a modified form of Debye and Hückel's theory of electrolytic conduction. McBain and Betz [1935] consider that the discrepancies are such as cannot always be explained by any reasonable modification of the existing theories.

III. INTERACTIONS OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS WITH NEUTRAL SALTS

When added to acid soils, neutral salts have long been known to liberate acid. Several well-known methods of estimating the lime requirement of soil, e.g. those due to Hopkins [1903], Daikuhara [1914] and Gedroiz [1924] are based on this principle. The nature of the interactions involved which has been the subject of controversies [Russell, 1937] is likely to be elucidated by systematic studies of the effect of neutral salts on hydrogen clays. Such studies have been made, comparatively recently by Wiegner [1931], Jenny [1932, 1936], Marshall and Gupta [1933], Mattson [1932, 1935] and others. The following investigations deal with some special features of the interaction.

1. *Interchanges between the diffusible H^+ ions associated with the colloidal particles of a hydrogen clay sol and the cations of an added salt*

In the following table the differences of the pH of hydrogen clay sols H and P and those of their ultrafiltrates have been compared with the corresponding differences observed when the sols contained sodium and potassium chlorides in small concentrations. -

** As C_{H^+} is of the order of $10^{-5}N$ to $10^{-4}N$, the concentration and the activity have the same significance.

* Discrepancies between the actual and calculated specific conductivities of hydrogen bentonite systems have been reported by Hauser and Reed (*J. Phys. Chem.* **41**, p. 911, 1937) since the communication to the Indian Science Congress Association of the paper dealing with the results given in this section. Similar discrepancies with hydrogen bentonites have also been observed by us (unpublished work).

TABLE IV

System	pH	e. m. f. (in volts) of conc. cell $\text{H}_2/\text{Sol} + \text{salt}/\text{sat. KCl}/$ $\text{ultrafiltrate}/\text{H}_2$ of I	
		I	II
		(Obs.)	(Calc.)
Sol H	4.52		
Ultrafiltrate of sol H	6.05		
Sol H + 0.0005N KCl	4.26		
Ultrafiltrate of above	4.42		
Sol H + 0.002N KCl	4.10		
Ultrafiltrate of above	4.15		
Sol P*	4.54		
Ultrafiltrate of sol P	5.85		
Sol P + 0.0005N NaCl	3.85	0.032	0.080
Ultrafiltrate of above	4.35		
Sol P + 0.002N NaCl	3.71	0.014	0.014
Ultrafiltrate of above	3.95		
Sol P + 0.0005N KCl	3.80	0.027	0.029
Ultrafiltrate of above	4.28		
Sol P + 0.002N KCl	3.70	0.011	0.012
Ultrafiltrate of above	3.90		

The differences between the pH values of the sol P + salt mixtures and those of the corresponding ultrafiltrates were checked by e. m. f. measurements with concentration cells of the type $\text{H}_2/\text{Sol} + \text{Salt}/\text{Sat. KCl}/\text{Ultrafiltrate of I}/\text{H}_2$.

I
II

The actual e. m. f.'s are in good agreement with those calculated from the pH values of I and II separately determined.

The results given in Table IV show that on the addition of the salt the pH of the sol itself changes only to a small extent compared to that of the ultrafiltrate. Apparently, a change has taken place in the sol in the location of the H^+ ions and their distribution between the colloidal particles and the intermicellary liquid which constitute two distinct phases. Hydrogen ions which were previously intercepted by the membrane can now pass through it. Originally, these hydrogen ions were associated with the colloidal particles themselves though mostly in an osmotically active, or, diffusible condition.

* This sol was prepared from a Deccan black soil (type 'B') kindly supplied by Dr J. K. Basu of the Sugarcane Research Station, Padegaon, Bombay.

On the addition of the neutral salt they have interchanged their positions with the cations of the salt and have passed into the intermicellary solution. This however, has not greatly affected the observed H^+ ion activity of the sol as the H^+ ions taking part in the interchange were mostly in an osmotically active, i.e. diffusible condition previous to the interchange. The marked increase of the H^+ ion activity of the ultrafiltrate, on the other hand, is readily explained by their displacement into the intermicellary solution. Similar observations with silicic acid sols have previously been made by Mr. B. Chatterjee in this laboratory (cited by Mukherjee, Mitra and Mukherjee [1937]).

2. *Interchanges between diffusible as well as non-diffusible H^+ ions associated with the colloidal particles of a hydrogen clay sol and the cations of an added salt*

The variations of pH of hydrogen clay sols H and P recorded in Table IV are small as the salt was added only in small concentrations. Considerably larger variations were observed using larger concentrations of salts as the following results will show :—

TABLE V

Sol	pH of sol	CH^+ of sol $\times 10^4$	Salt added and conc. of salt	pH of sol + salt	CH^+ of Sol + salt $\times 10^4$
E	4.66	0.22	0.10 N NaCl	3.61	2.45
			0.10 N $CaCl_2$	3.52	3.02
			0.10 N $BaCl_2$	3.15	7.08
H	4.52	0.32	0.80 N NaCl	3.46	3.46
			0.80 N $CaCl_2$	3.33	4.67
			0.80 N $BaCl_2$	3.23	5.87
I	4.51	0.31	0.25 N $CaCl_2$	3.43	3.72
			0.25 N $BaCl_2$	3.31	4.90
K	4.47	0.34	0.25 N $CaCl_2$	3.30	5.01
			0.25 N $BaCl_2$	3.02	9.55

The curves given in Figs. 1 and 2 show the variations in the H^+ ion activity of sols E and H on progressive additions of NaCl, $BaCl_2$ and $CaCl_2$.

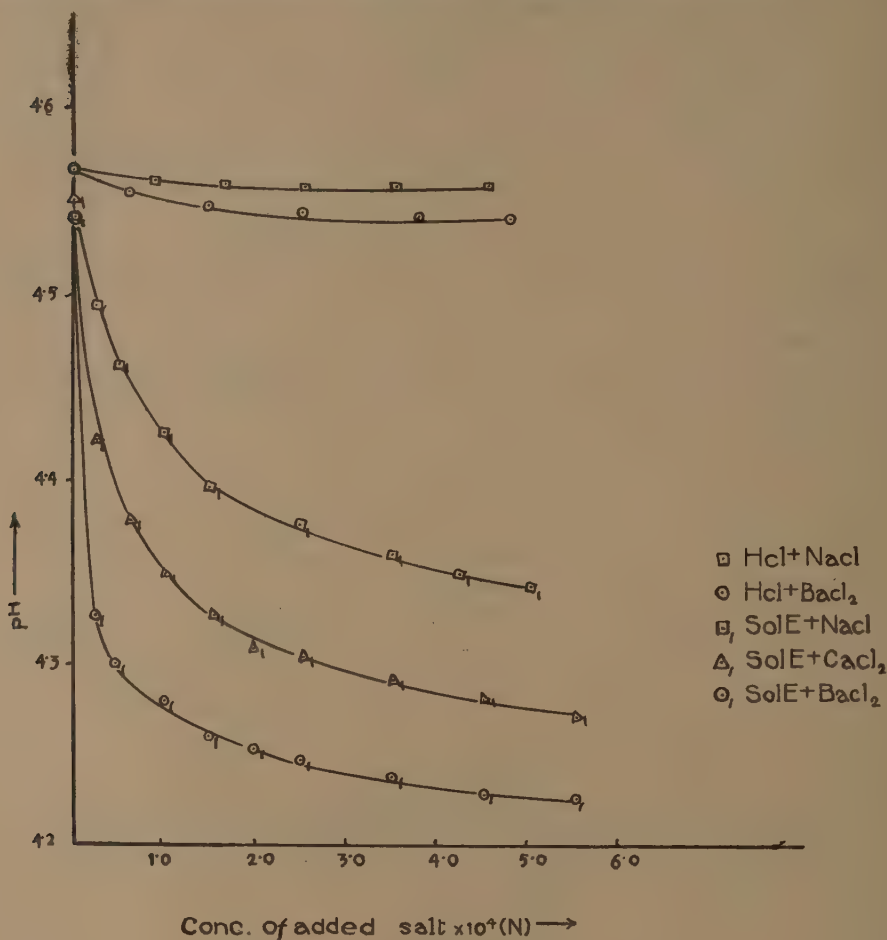


FIG. 1

The curves as also the results recorded in Table V show, in agreement with previous observations [Mitra, 1936], that using chlorides alone, the effect of different cations in increasing the H^+ ion activity is in the order $Ba^{++} > Ca^{++} > Na^+$ which follows the usual lyotrope series. The increase of the H^+ ion activity may be due (i) to an alteration of the activity coefficient of the free (osmotically active) hydrogen ions associated with the colloidal particles and/or (ii) to a change from an osmotically inactive to an active condition of H^+ ions following the addition of the salt. Without the salt, these H^+ ions (ii) do not register their activity on the electrode. The activity of free H^+ ions in true solutions of acids would not be altered to any such extent on the addition of the salt. This is shown by the control experiment on the H^+ ion activity of a hydrochloric acid solution having nearly the same pH as the sol (Fig. 1).

Taken in conjunction with the ultrafiltration experiments on sols H and P the observed large variations of H^+ ion activity recorded in Table V arise at least in part from a displacement of osmotically inactive H^+ ions from the colloidal particles.

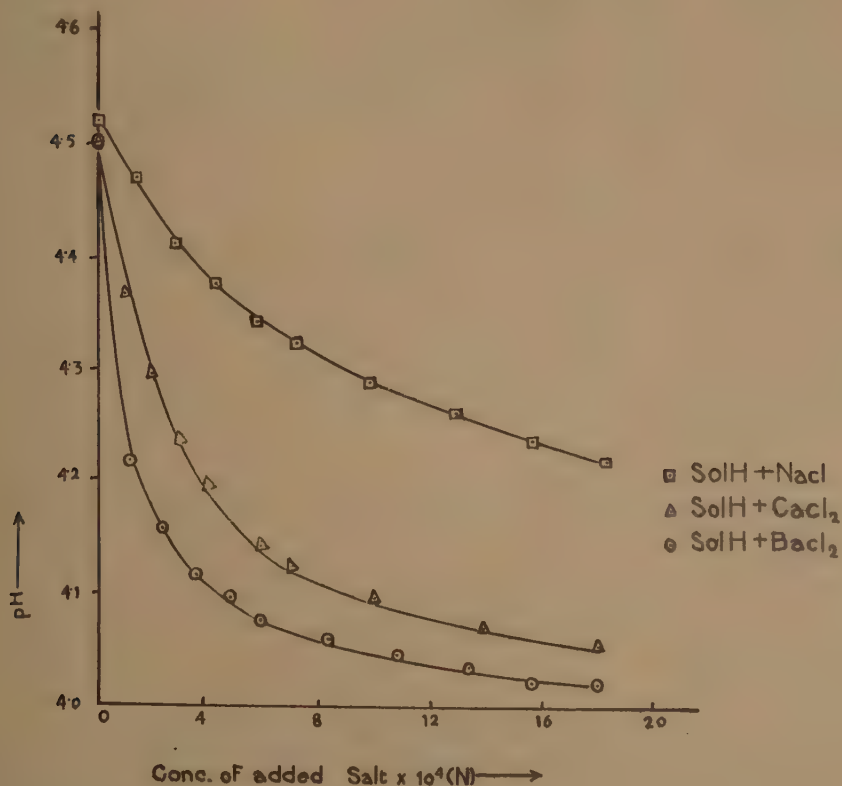


FIG. 2

Aluminium ions displaced from hydrogen clays and acid soils by neutral salts are known to contribute to the observed free and total acids of the neutral salt extracts* [Paver and Marshall, 1934]. The following results show that such aluminium ions cannot wholly account for the observed free and total acids of the supernatant liquids above the coagula of hydrogen clay sol + salt mixtures.

An exchange of osmotically inactive H^+ ions associated with the colloidal particles of the sols for the cations of the salt has to be assumed in order to explain the much larger free and total acids of the ultrafiltrates of the sol + salt mixtures compared to those of the ultrafiltrate of the sol itself.

* This topic is being systematically studied by Mr B. Chatterjee in this laboratory.

TABLE VI

System	pH	Free acid (gm. ions of H per litre)	Total acid at inflexion point of titration curve with NaOH		Al in ultra- filtrate (milli-equi- valents per 100 gm. colloid)
			1 Equivalents per litre	1 Millie-qui- valents per 100 gm. colloid	
Sol P	4.54	2.9×10^{-5}	200×10^{-5}	82.0	..
Ultrafiltrate of sol P	5.85	1.4×10^{-6}	<i>Nil</i>	<i>Nil</i>	..
Ultrafiltrate of sol P + .01N NaCl (curve I, figure 2 a).	3.66	2.2×10^{-4}	2.4×10^{-4}	10	0.5
Ultrafiltrate of sol P + .04N NaCl (curve II, figure 2a).	3.30	5.0×10^{-4}	4.6×10^{-4}	19	3.0
Solution of alumi- nium chloride* (curve III, Fig. 2a).	3.22	6.0×10^{-4}	1.26×10^{-1}

The titration curves of the ultrafiltrates of the sol+salt mixture given in Fig. 2(a) are characteristic of a dissolved strong acid, viz., hydrochloric acid, formed by the interaction between the hydrogen clay and the neutral salt, sodium chloride.

The curves have a different form compared with that of a solution of aluminium chloride given in the same figure. The strong acid character of the titration curves of the ultrafiltrates harmonizes with the result (Table VI) that they have nearly the same free and total acids expressed in normality. The solution of aluminium chloride, on the other hand, has very different free and total acids.

Table VI shows that at the higher concentration of the salt the displaced aluminium forms a larger part of the total acid of the ultrafiltrate. Similar observations have been made by Paver and Marshall [1934] and by B. Chatterjee in this laboratory.† They will be fully dealt with in a future publication.

* From unpublished work by Mr N. P. Datta in this laboratory

† Unpublished work

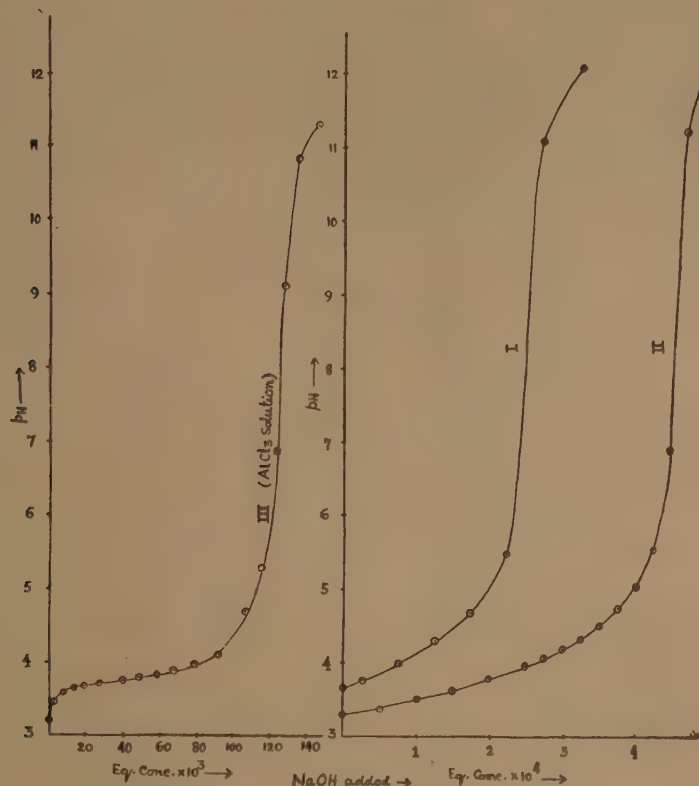
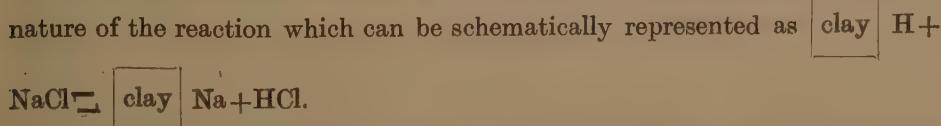


FIG. 2(a)

The following results obtained with sol P show that by repeated treatment of the sol with a solution of sodium chloride at a sufficiently low concentration ($0.005N$), increasing amounts of H^+ ions can be displaced from the colloidal particles of the sol though practically no Al^{+++} ions are found in the salt extracts.

Theoretically it is not impossible that if the leaching is sufficiently continued an amount of H^+ ions comparable to the total acid of the sol calculated at the inflexion point of its titration curve with a base (e.g. $NaOH$) will be displaced from the colloidal particles. The incomplete displacement of the H^+ ions by the cation of the salt is obviously to be referred to the balanced nature of the reaction which can be schematically represented as



The incomplete displacement is due to the back reaction whose intensity at any stage of the leaching depends on the concentration of free H^+ ions in

the intermicellary liquid. This concentration decreases as the leaching proceeds thus favouring more and more the direct reaction. In the interaction with a base to be discussed later the back reaction is absent thus securing a more complete replacement of the H^+ ions of the colloidal particles by the cations of the base though the cations may be at a much lower concentration than in the case of the neutral salt solution.

TABLE VII

System	Total acid in m. e. per 100 gm. colloid*
Sol P	82.0
1st leachate (0.5 gm. of clay + 200 c.c. of 0.005N NaCl solution)	9.0
2nd leachate (50 c.c. solution)	2.0
3rd leachate ,, 	1.8
4th leachate ,, 	2.0
5th leachate ,, 	1.8
6th leachate ,, 	2.0

* Calculated at inflexion point of potentiometric titration curve with NaOH.

IV. INTERACTIONS OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS WITH BASES : THE NATURE OF THE TITRATION CURVES

In part V of this series [Mitra, 1936] the nature of the potentiometric titration curves of a number of hydrogen clay sols with bases has been discussed. In this paper, both potentiometric and conductometric titration curves obtained under diverse conditions of titration have been examined including detailed analyses of their slopes and buffer capacities in different pH regions. Such systematic studies are likely to throw considerable light on the nature of the acid-base interaction in soil. They have been seldom undertaken in the past. Reference may, however, be made to Knight [1920], Jensen [1929], Hardy and Lewis [1929] and others who obtained titration curves

of soils mainly in connexion with the estimation of the lime requirement, or the degree of saturation of soil.

1. Potentiometric titration curves of hydrogen clay sols with different strong bases

Figs. 3*, 4, and 5 give the titration curves of hydrogen clay sols E, G and H obtained on titrating them with sodium, barium and calcium hydroxides.

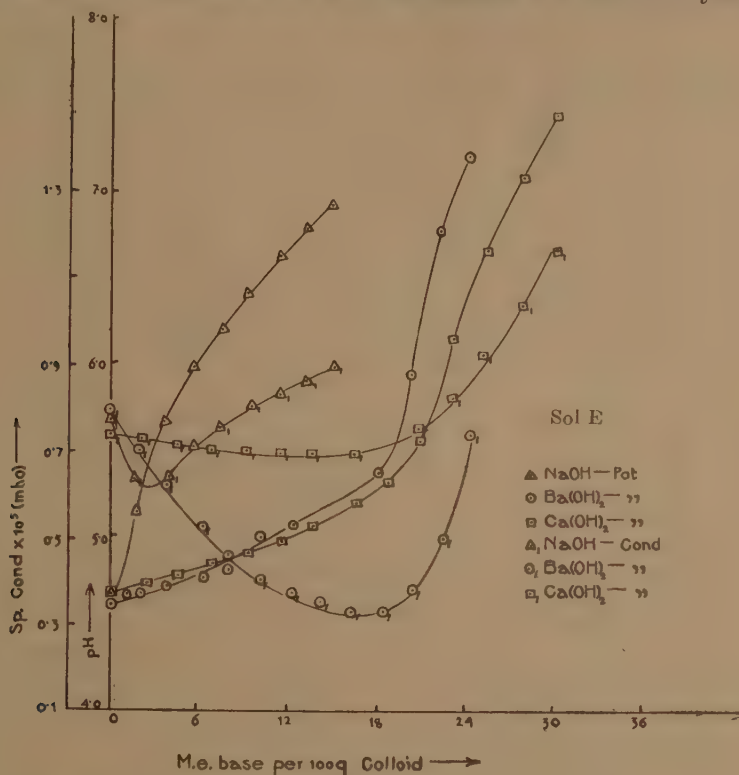


FIG. 3

The forms of the curves with the different bases are different. The potentiometric titration curves with baryta and calcium hydroxide indicate an apparent strong monobasic acid character of the sols. There is an initial flat portion followed by a steeper region which shows an inflexion point. The slope gradually diminishes after the inflexion point has been passed. Bayer [1930] observed an initial sharp rise in the potentiometric titration curves of hydrogen clay sols with the above bases and from this observation he attributed a weak monobasic acid character to the sols studied by him [cp. Bradfield, 1923].

The strong monobasic acid character of the potentiometric titration curves with barium and calcium hydroxides cannot be referred to the neutralization of any dissolved acid or acids present in the sol as its ultrafiltrate contains

*Rep oduced from Fig. 1 of part V of this series,

negligible free and total acids (Table II). A difference from a true monobasic acid character, however, exists. Thus the free acidity of sol G, for example, is only 6.5 per cent of its total acid even at a total acid concentration of the order of $10^{-4}N$. On analogy with an acid in true solution, the sol is very weakly dissociated. The potentiometric titration curve of the sol with baryta, however, (Fig. 4), does not show the sharp initial rise characteristic of a weak acid in true solution.

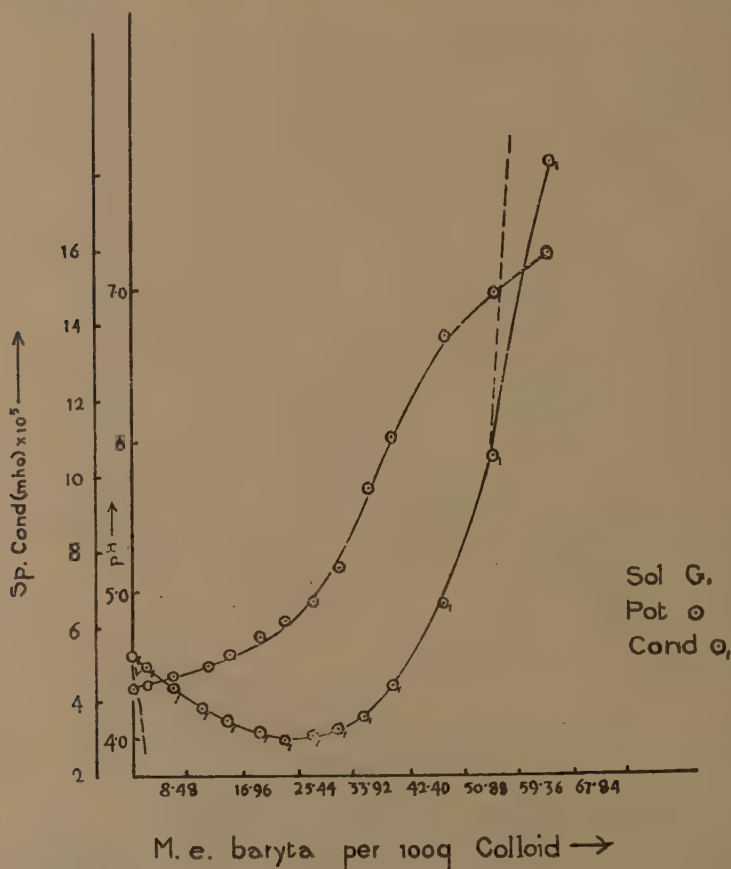


FIG. 4

The potentiometric titration curves with caustic soda show a comparatively sharp initial rise indicating a relatively weak acid character of the sols. This initial rise is followed by a gradual flattening of the curve, that is by a region of continued and increasing buffer action. No inflexion point in the alkaline region characteristic of a weak acid in true solution is, however, observed in the curves of sols E and H up to the point to which the titration had been extended. There is thus an important difference from a true weak acid character although a consideration of the comparatively sharp initial rise alone would justify the conclusion that a weak acid is being titrated,

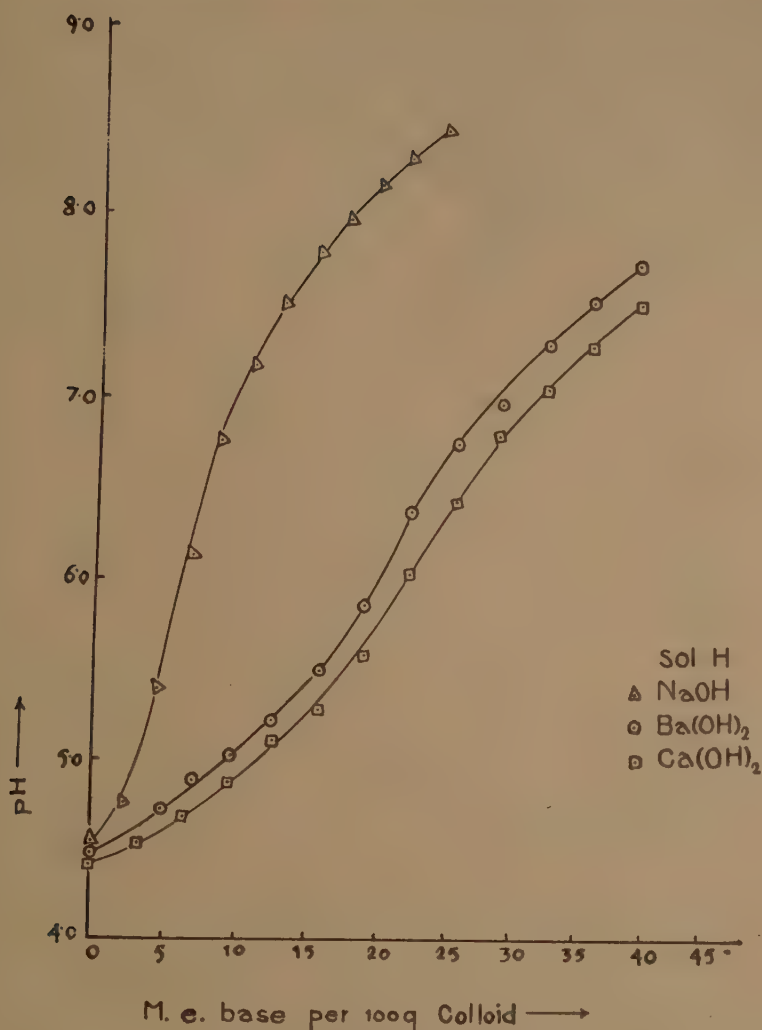


FIG. 5

Compared to sols E and H, the caustic soda titration curves of sols I and K given in Figs. 6 and 7 closely resemble those of a weak acid in true solution in that they have definite inflexion points in the alkaline region.

Sols I and K differ from sols E and H in one important respect. The silica-alumina ratios of their colloidal material are considerably greater than those of sols E and H as the following figures will show.

TABLE VIII

SiO ₂ /Al ₂ O ₃ (molar)	Colloidal material of sol			
	E	H	I	K
	2.65	2.37	3.87	3.40

A closer examination of the slopes of the caustic soda titration curves of sols I and K and of their buffer capacities show, further, that the weak monobasic acid character indicated by them is only apparent. Thus the pH at the point of half neutralization in the caustic soda titration curve of sol I is 6.05 which corresponds to a dissociation constant of 8.91×10^{-7} . In Fig. 8 the theoretical titration curve of a monobasic acid having this dissociation constant and the total acidity corresponding to the inflexion point of sol I has also been given for comparison.

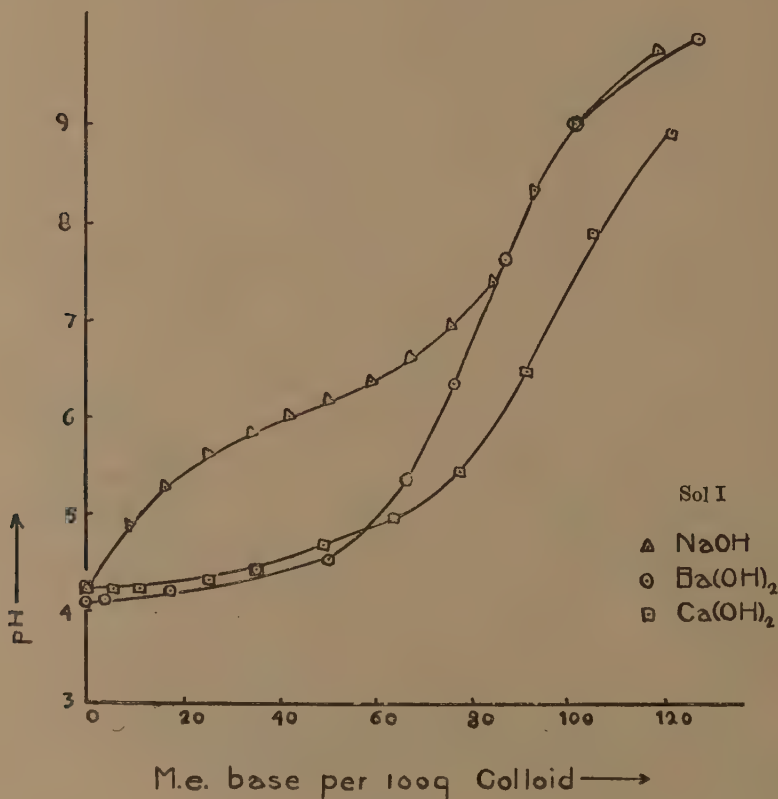


FIG. 6

The theoretical curve has been obtained with the aid of the equation

$$[H] = K \cdot \frac{a - b - [H^+] + [\overline{OH}]}{b + [H^+] - [\overline{OH}]}$$

where a represents the total acid, b the concentration of the base and K the dissociation constant. The theoretical titration curve definitely differs from the actual titration curve. A comparison of the buffer capacities* which are

*The buffer capacity considered here is the reciprocal of the slope of the potentiometric titration curve and is given by the expression (van Slyke : *J. Biol. Chem.*, 22, p. 525, 1922) $\frac{2.302 \cdot a \cdot K \cdot [H^+]}{(K + [H])^2}$ where (a) is the total acidity and K , the dissociation constant,

also shown in Fig. 8 confirms the difference between the two curves specially after the inflexion point has been passed. The sol has a higher buffer capacity than the corresponding hypothetical acid. It is evident that the sol has definitely different properties than what are expected of a corresponding weak monobasic acid in true solution.

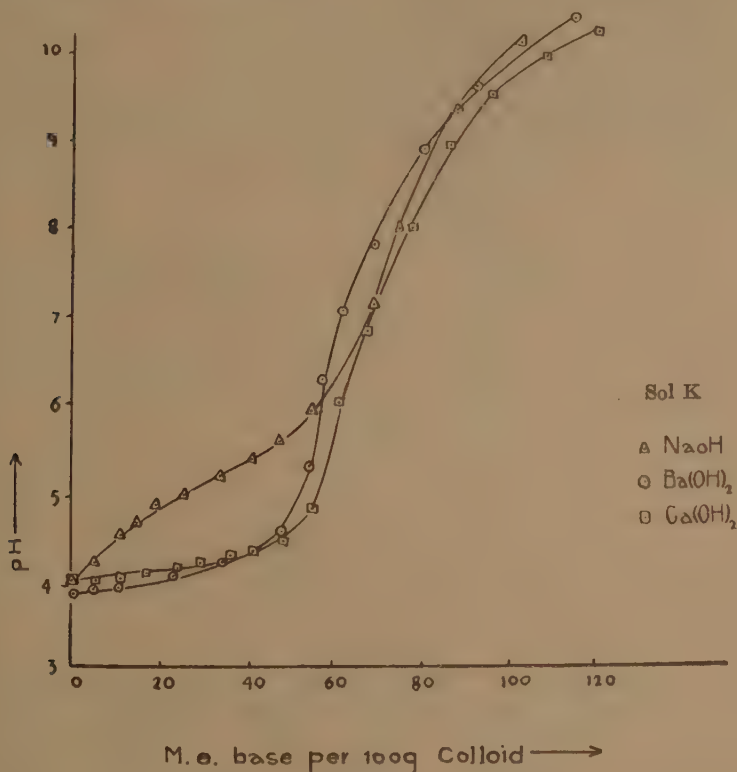


FIG. 7

The baryta and calcium hydroxide titration curves of sols I and K show, as in the case of sols E and H, an apparent strong monobasic acid character.

The titration curves with the different bases yield different total acidity values calculated from their inflexion points. These variations of the total acid have been discussed in detail in the previous paper of this series. Attention should, however, be drawn here to an interesting difference between sols E and H on the one hand and sols I and K on the other, in respect of the order of total acidities obtained on titration with different bases. With sols E and H, the total acid follows the order : $\text{Ca}(\text{OH})_2 > \text{Ba}(\text{OH})_2 > \text{NaOH}$. The NaOH titration curves of sols I and K, however, give total acidity values (calculated at the inflexion points) which are somewhat greater than those obtained from their baryta and calcium hydroxide titration curves. This is brought out by Table IX.

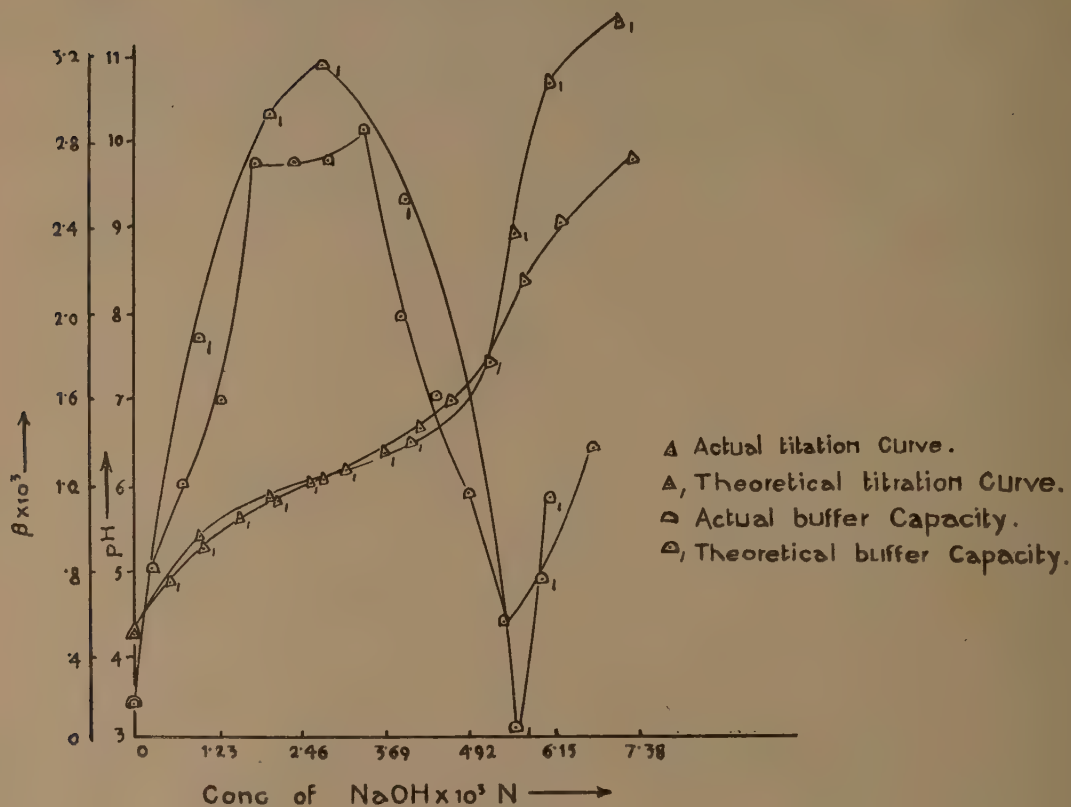


FIG. 8

TABLE IX

Sol	Base used for titration	pH at inflexion	Total acid in m.e. base per 100 gm. colloid	
			At inflexion pt.	At pH 7.0
E	NaOH	5.4	2.2	15.4
	$\text{Ba}(\text{OH})_2$	6.0	20.6	25.0
	$\text{Ca}(\text{OH})_2$	5.8	21.5	26.2
H	$\text{Ba}(\text{OH})_2$	5.8	21.5	32.0
	$\text{Ca}(\text{OH})_2$	6.6	21.5	32.8
I	NaOH	8.05	90.0	78.0
	$\text{Ba}(\text{OH})_2$	7.00	82.0	82.0
	$\text{Ca}(\text{OH})_2$	6.95	96.0	97.0
K	NaOH	7.15	68.0	67.0
	$\text{Ba}(\text{OH})_2$	5.80	55.0	61.0
	$\text{Ca}(\text{OH})_2$	5.20	58.0	67.0

In comparing the total acid at the inflexion points in the titration curves with different bases, the location in the pH scale of such inflexion points has to be considered for, as was shown in the previous part of this series, the higher the pH the greater is the amount of the acid reacting with the base. The greater total acid (at the inflexion point) with NaOH than with Ba(OH)_2 , or, Ca(OH)_2 observed in the case of sols I and K is due to the inflexion points in the NaOH curve occurring in the alkaline region which is not the case with the Ba(OH)_2 , or, Ca(OH)_2 curves. The titration curves of sols E and H do not show such features. An examination of the slopes of the titration curves of sols I and K with different bases shows the same order of the capacity of these bases to react with sols I and K as observed with sols E and H. The order is : $\text{Ca(OH)}_2 > \text{Ba(OH)}_2 > \text{NaOH}$.

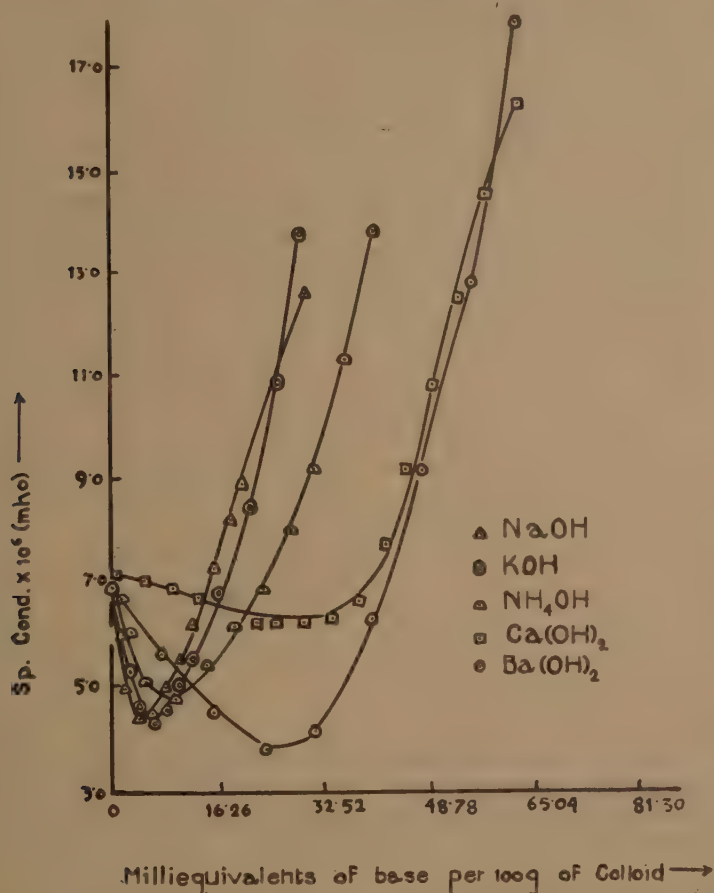


FIG 9.

2. Mutually conflicting features of the potentiometric and conductometric titration curves of hydrogen clay sols.

The conductometric titration curves of hydrogen clay sols with different bases present certain features which are entirely at variance with those of the

corresponding potentiometric titration curves. No reference to such discrepancies is found in the existing literature.

The potentiometric titration curve of sol E with caustic soda shows an initial weak acid character while the corresponding conductometric curve (Fig. 3) has a sharp minimum (more sharp than the minimum of the baryta or calcium hydroxide titration curve) and in this respect indicates a strong acid character of the sol. Bayer [1930], however, observed an initial rise of the specific conductivity on titrating a hydrogen clay sol with caustic soda. This was obviously due to too much alkali being added at the very first instalment, the initial lowering of the specific conductivity and the minimum point being thus missed.

The conductometric titration curves of sols E and H with baryta and calcium hydroxide have round or flat minima and in this respect indicate, in contrast to the corresponding potentiometric titration curves, a comparatively weak acid character of the sols.

Fig. 9 offers an interesting comparative study of the conductometric titration curves of hydrogen clay sol F with sodium, potassium, ammonium, calcium and barium hydroxides.

The slopes of the descending and part of the ascending portion of the curve obtained on titrating with $\text{Ca}(\text{OH})_2$ is distinctly less than that of the curve with $\text{Ba}(\text{OH})_2$. The slopes are arranged in the order : $\text{NaOH} > \text{KOH} > \text{NH}_4\text{OH} > \text{Ba}(\text{OH})_2 > \text{Ca}(\text{OH})_2$ as the following figures will show :

TABLE X

Base used	Slope of initial descending portion	
	obs.	Calc.
NaOH	0.100	0.346
KOH	0.085	0.318
NH_4OH	0.045	0.315
$\text{Ba}(\text{OH})_2$	0.018	0.330
$\text{Ca}(\text{OH})_2$	0.003	0.335

The NaOH curve has the greatest downward slope and in this respect shows a stronger acid character than the $\text{Ba}(\text{OH})_2$, or, the $\text{Ca}(\text{OH})_2$ curve. As already shown, however, the potentiometric titration curves with these bases give an altogether different picture regarding the acid character of the sols.

Table X shows that the actual slopes of the descending portions of the conductometric curves are smaller than those calculated* for a strong acid.

* The slope is given by $(U_{\text{H}^+} - U_{\text{M}})/1000$, where U_{H^+} and U_{M} are respectively the mobilities of hydrogen ion and the cation of the base at 35°C. at which all measurements were carried out.

The greatest discrepancy is observed with the $\text{Ba}(\text{OH})_2$ and $\text{Ca}(\text{OH})_2$ curves.

3. The variability of the total neutralizable acid of colloidal solutions of hydrogen clays

Reference has already been made to variations of the total acid of a hydrogen clay sol obtained on titration with different bases. In part VI of this series [Mitra, Mukherjee and Bagchi, 1940] these variations of the total acid have been fully reported and it has been shown that titration of the sol with a given base in the presence of a large concentration of a neutral salt yields a higher total acid, measured at the same $p\text{H}$, than titration with the base alone. Such variations would not be possible in the case of any dissolved acid.

V. THE 'DEGREES OF DISSOCIATION' AND 'DISSOCIATION CONSTANTS' OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS

If colloidal solutions of hydrogen clays could be treated as acids in true solution their degrees of dissociation at a given total acid concentration and dissociation constants calculated from their titration curves would give an estimate of their 'strength' as acid systems.

The degree of dissociation of a hydrogen clay sol at a given total acid concentration may be taken for purposes of discussion as equal to the ratio of the free acidity (i.e. the hydrogen ion concentration calculated from the observed $p\text{H}$) to the amount of acid equivalent to that of a dilute base necessary to reach the inflexion point. The following table gives degrees of dissociation of hydrogen clay sols E, F and H.

TABLE XI

Sol	Base used for the titration	$p\text{H}$	H ion conc. $\times 10^5 N$	Total acid at inflexion point $\times 10^5 N$	Free acidity $\times 100$
					Total acidity
E	NaOH . . .	4.66	2.19	2.6	84.2
	$\text{Ba}(\text{OH})_2$. . .			24.3	9.0
	$\text{Ca}(\text{OH})_2$. . .			25.0	8.4
F	NaOH . . .	14.4	3.89	12.0	32.4
	$\text{Ba}(\text{OH})_2$. . .			38.0	10.1
H	NaOH . . .	4.52	3.02	14.0	75.00
	$\text{Ba}(\text{OH})_2$. . .			99.0	3.03
	$\text{Ca}(\text{OH})_2$. . .			99.0	3.03

The degree of dissociation has a surprisingly low value when baryta or calcium hydroxide is used for the titration although the total acid concentration is of the order of $10^{-4} N$. The sol thus behaves as a very weak acid. The corresponding potentiometric titration curves indicate, on the other hand, a strong acid character of the sols as already pointed out.

Table XI shows, however, that much higher values of the degree of dissociation are obtained when the total acid given by the inflexion point, or minimum of the caustic soda titration curve, is used for the calculation. This is in agreement with the strong acid character of the conductometric titration curves with caustic soda (Fig. 4) but is at variance with the comparatively weak acid character of the corresponding potentiometric titration curve. The differences in the values of the degree of dissociation arise obviously from the variations of the total acidity of the sols previously mentioned.

The apparent dissociation constants of hydrogen clay sols E, I and K have also been calculated. Two series of values of the dissociation constant have been obtained and tabulated below as K and K' values. The K values were calculated from the equation $K = \frac{\alpha_c^2}{1-\alpha}$ where α is the ratio of the free acid to the total acid (C). The K' values were calculated from different points in the potentiometric titration curves using the equation $pH = pK' + \log \frac{[\text{salt}]}{[\text{acid}]}$; $[\text{salt}]$, at any stage, has been taken as equivalent to the concentration $[B]$ of the base added and $[\text{acid}]$ has been taken as equal to $C - [B]$.

TABLE XII

Sol	Base used for titration	K	K'		
			1/4 neutralization	1/2 neutralization	3/4 neutralization
E	Ba(OH) ₂ .	2.2×10^{-6}	1.4×10^{-5}	2.8×10^{-5}	5.0×10^{-5}
	Ca(OH) ₂ .	2.0×10^{-6}	5.0×10^{-6}	1.0×10^{-5}	1.7×10^{-5}

TABLE XIII

Sol	Base used for titration	K	K'		
			1/4 neutralization	1/2 neutralization	3/4 neutralization
I	NaOH .	7.3×10^{-7}	8.9×10^{-7}	7.9×10^{-7}	8.0×10^{-6}
	Ba(OH) ₂ .	8.9×10^{-7}	2.5×10^{-5}	3.7×10^{-5}	3.5×10^{-5}
	Ca(OH) ₂ .	6.5×10^{-7}	2.2×10^{-5}	2.4×10^{-5}	2.2×10^{-5}
K	NaOH .	6.7×10^{-7}	5.6×10^{-6}	5.3×10^{-6}	4.2×10^{-6}
	Ba(OH) ₂ .	8.9×10^{-7}	4.4×10^{-5}	7.0×10^{-5}	1.0×10^{-4}
	Ca(OH) ₂ .	8.5×10^{-7}	3.9×10^{-5}	6.3×10^{-5}	1.0×10^{-4}

Tables XII and XIII show that there is no agreement between the K and K' values especially for the baryta and calcium hydroxide titrations. Also, the values of K' for them are much larger than those for the caustic soda titrations. This is again in agreement with the strong acid character of the baryta and calcium hydroxide titration curves (potentiometric) compared to the weak acid character of the caustic soda curves (potentiometric). Actually, the 'dissociation constants' given in tables XII and XIII are not constants in any real sense of the term and consequently they lose their significance.

VI. THE ROLE OF THE ELECTRICAL DOUBLE LAYER AND OF ADSORPTION OF IONS IN THE INTERACTIONS OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS WITH BASES AND SALTS

The results given in the preceding sections show that the interactions of hydrogen clay sols with electrolytes present a number of special features which are difficult to reconcile in the light of classical electrochemistry. A much clearer elucidation of these special features can, however, be obtained on the basis of the theory of the electrical double layer postulating the existence of primarily adsorbed ions associated with the colloidal particles of the sol and of a secondary adsorption of cations by them [Mukherjee, 1921, 1922]. Mukherjee, Mitra and Mukherjee [1937] have used this theory as the basis of a theoretical formulation of the interactions of colloidal acid systems including hydrogen clay sols. An outline of the theory has been given in the previous paper of this series [Mitra, Mukherjee, and Bagebi, 1940] where it has been applied to explain the variations of the total neutralizable acids of hydrogen clay sols observed on estimating the acid under different conditions of titration. An explanation of the special features of hydrogen clay sols recorded in this paper is given below based on the above theory.*

According to the theory, H^+ ions corresponding to the primarily adsorbed anions 'built in' on the solid side of the solid-liquid interface exist in two states, viz. in a secondarily adsorbed condition either by electrostatic, or, specific forces and in a free, or, 'mobile' state. The H^+ ions of the first category are osmotically inactive. They are the 'bound' H^+ ions. Both 'mobile' and 'bound' H^+ ions may be displaced by the cations of an added salt, or, a base, the displacement being determined by the adsorbability of the cations given by their valency, mobility and state of hydration when the cations are adsorbed by electrostatic forces of attraction. They may also be adsorbed by specific valence forces, or forces of the Van der Waals' type.

The mobile H^+ ions give rise to the free acidity, i.e. the observed H^+ ion activity of the sol while its total acidity calculated from the inflexion point of its titration curve with a base includes both 'mobile' and 'bound' H^+ ions. With the sols used in this work, the 'bound' H^+ ions far outnumber the ions of the other category as the small ratio of the free to total acids of the sols shows.

*The picture here suggested is of a general nature and it takes no account of (a) the detailed mineralogical structure of hydrogen clays, (b) their amphoteric character and (c) the role of Al^{+++} and other ions on the surface in addition to H^+ ions. Investigations covering these aspects are in progress.

Though osmotically active, the 'mobile' H^+ ions are not present in the intermicellar liquid of the sol which explains the negligible free and total acids of the ultrafiltrate of the sol compared to those of the sol itself. The colloidal particles of the sol with their adsorbed H^+ ions—mobile and bound—constitute a distinctly separate phase from the intermicellar liquid.

On the addition of a neutral salt to the sol, only the mobile H^+ ions, or both mobile and bound H^+ ions, may be displaced by its cations. The alkali metal cations are weakly adsorbed. Consequently, when a salt containing an alkali metal cation is added to the sol, a displacement of the mobile H^+ ions only may take place specially if the salt is added in very low concentrations. As only a displacement of hydrogen ions which were previously in an osmotically active condition is involved, no marked variation of the hydrogen ion activity of the sol will be observed. The results given in table IV illustrate this effect. Though the H^+ ion activity of the sol has not appreciably changed, that of its ultrafiltrate has considerably increased. This increase is the result of the interchange between the mobile H^+ ions in the double layers and the alkali metal cations in solution.

A displacement of the bound H^+ ions will cause an increase in the H^+ ion activity of the sol. The results given in table V show that using chlorides alone, the relative effects of the different cations in increasing the H^+ ion activity follows the order $Ba^{++} > Ca^{++} > Na^+$ which is the order of their electrical adsorption. In the previous paper of this series [Mitra, Mukherjee, and Bagchi, 1940] it has been shown that the relative effects of the cations to increase the total neutralizable acid of the sol follow the same order which is also in agreement with the usual lyotrope series. It appears, therefore, that in the interactions of hydrogen clays with neutral salts an electrical adsorption of the cations of the salt plays a dominant role. The resulting cation effect is thus determined by electrical factors alone and it may consequently be designated as the '*regular cation effect*'.

In the interactions of hydrogen clays with bases also, adsorption of the cations of the base plays a definite role. On the addition of a base, besides the direct neutralization of the H^+ by the OH^- ions, the cations of the base displace some of the bound H^+ ions from the double layer which are then neutralised by the OH^- ions. The greater the adsorbability of the cation, the greater is this displacement and hence a larger amount of acid is neutralized at a given pH. The smaller total acidity obtained on titration with sodium hydroxide compared to barium and calcium hydroxides is thus explained. The total acid calculated at a fixed pH decreases in the order $Ca(OH)_2 > Ba(OH)_2 > NaOH$. In the interactions of the sols with the bases, therefore, the Ca^{++} ions appear to have a greater relative effect than the Ba^{++} ions. The slopes of the titration curves point to the same relative effects of the cations. Here, therefore, we have an '*irregular*' or '*specific cation effect*' in the sense that it does not follow the lyotrope series. Unlike the regular cation effect previously discussed it does not result from simple electrical adsorption of the cations together with their hydrated envelopes but arises from their adsorption in the dehydrated condition by specific forces other than simple electrostatic forces.

The features of the titration curves are explained from similar considerations. The first additions of the base neutralize the mobile H^+ ions. The disappearance of these mobile H^+ ions displaces the equilibrium between mobile and bound H^+ ions in the double layer which is restored by the passage of some bound H^+ ions from the bound to the mobile condition. Adsorption of the cations of the base considerably facilitates this process. When barium or calcium hydroxide is the base used for the titration, the Ba^{++} or Ca^{++} ions, because of their high adsorption, displace more and more bound H^+ ions from the beginning of the titration which are then neutralized by the OH^- ions of the base. The titration curve (potentiometric) has, therefore, a flat run indicating a moderately strong acid character of the sol. When the limit to which the bound H^+ ions can be so displaced and neutralized has been reached, further addition of the base results in a sharp rise of the pH , that is, the titration curve shows an inflexion point. This limit, however, does not correspond to the neutralization of all the bound H^+ ions as the titration curve shows a continued buffer action beyond the inflexion point. The inflexion point thus indicates the neutralization of H^+ ions in a definite affinity level.

Using sodium hydroxide also, the first additions of the base neutralize the mobile H^+ ions. The bound H^+ ions which far outnumber the mobile H^+ ions cannot be displaced from the double layer by the sodium ions because of their weak electrical adsorption. The pH of the system, therefore, shoots up and the titration curve shows a comparatively sharp initial rise. On further additions of the base, the concentration of sodium ions in the system increases and thus the probability of their adsorption is increased. This, combined with the gradually increasing pH of the system helps in the neutralization of more and more bound H^+ ions and the titration curve shows a flattening after the initial rise. When the limit to which the bound H^+ ions can be so displaced and neutralized has been reached further addition of the base may result in a sharp rise of the pH , that is, an inflection point in the titration curve may be observed (titration curves of sols I and K in Figs. 6 and 7).

A consistent explanation of the apparently contradictory features of the potentiometric and conductometric curves is also obtained on the assumption that the greater the electrical adsorbability of the cations of the base the greater is the amount of bound H^+ ions displaced from the double layer which can then react with the OH^- ions of the base. The greater the displacement of bound H^+ ions the smaller will be the slopes of the conductometric titration curves which will thus resemble those of a weak acid. The marked departure in the slopes of the $Ca(OH)_2$ and $Ba(OH)_2$ curves of sol F from those calculated for a strong acid and the weak acid character of these curves are thus explained. The process of displacement of bound H^+ ions and their subsequent neutralization would also diminish the slope of the potentiometric titration curve but in this case a smaller slope indicates a stronger acid. The caustic soda titration curve (conductometric) has the greatest downward slope though the corresponding potentiometric curve shows the steepest initial rise and thus the weakest acid character. The total acid calculated from the minimum of the $NaOH$ curve agrees nearly (within 15 per cent) with the free

acidity of the sol. The minimum of the caustic soda curve thus corresponds mainly to the neutralization of the mobile H^+ ions and only a small fraction of the bound H^+ ions.

Summary

The electrochemical properties of a number of hydrogen clay sols have been studied.

While the sols have measurable free and total acids, their ultrafiltrates are practically neutral.

The actual specific conductivity of the sol is often less than that due to its free H^+ ions whose concentration has been calculated from the observed pH of the sol.

When a neutral salt is added to a hydrogen clay sol, its H^+ ion activity shows a marked increase. The nature and concentration of the cations of the salt are important factors. Using chlorides alone, the relative effects of cations follow the order $Ba^{++} > Ca^{++} > Na$ which is in agreement with the lyotrope series and thus illustrates a 'regular cation effect'.

The sols give characteristic potentiometric and conductometric titration curves on titration with bases. The curves show several features which would not be expected with dissolved acids. The forms of the curves obtained on titration with different bases as also the total neutralizable acids of the sols calculated from them are different. The total acid decreases in the order $Ca(OH)_2 > Ba(OH)_2 > NaOH$. The greater relative effect of $Ca(OH)_2$ compared to $Ba(OH)_2$ illustrates an 'irregular, or, specific cation effect'. Mutually conflicting features are shown by the potentiometric and conductometric titration curves with a given base. The 'dissociation constants' calculated from the potentiometric curves have a fictitious significance. Somewhat different types of curves are obtained with hydrogen clays having widely different silica-alumina ratios.

The results have been discussed from the point of view of classical electrochemistry as also in the light of the theory of the electrical double layer and of adsorption of ions.

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THE BASE BINDING CAPACITIES OF HYDROGEN CLAYS AS DETERMINED BY DIFFERENT METHODS, I*

BY

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THERE is an element of arbitrariness regarding the various routine methods for estimating the base binding, or, base exchange capacity of soil. The different alternative methods do not always give concordant results. The uncertainty mainly arises from the difficulty of an accurate and unequivocal definition of what constitutes the exchangeable hydrogen, or, the titratable acid of the soil. Unlike the estimation of acids in true solution, or, colloidal systems in which the various phases can be clearly defined the amounts of acids estimated by the different routine methods are often ill-defined. A precise knowledge of the nature of the interactions involved in the estimations is therefore desirable especially in order to render possible a satisfactory correlation of the mutually conflicting experimental observations.

The methods used by Hopkins [1903], Daikuhara [1914] and Gedroiz [1924] for the estimation of the lime requirement of soil are based on the liberation of acid by the interaction of an acid soil with a neutral salt. Several theories have been proposed to explain the nature of this interaction. Until recently, this interaction had been regarded by some [Joseph and Oakley, 1925], following Way [1852], to be an instance of double decomposition. It has also been suggested that on account of surface tension effects [Gedroiz, 1929] the neutral salt is split into the acid and the base of which the base is adsorbed at the interface and the corresponding acid is liberated. The difficulties in the way of a simple explanation is illustrated by observations such as those of Ramann [quoted by Hissink, 1935] that a complete displacement of all the reactive hydrogen ions of soil is not effected even by continued leaching of the soil with a salt solution. The problem is further complicated by the fact that aluminium ions are nearly always found in the neutral salt extracts of acid

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soils. The role played by the electrical double layer in these interactions has been emphasized by Mukherjee [1922] and Wiegner [1925]. The picture is however still far from being definite and according to Hissink [1935], no existing theory can adequately explain the nature of the above interactions.

The method of electrometric titration with a base has often been used [Goy, Muller and Roos, 1928 ; Hardy and Lewis, 1929] for estimating the total neutralizable acid of soil, e.g. for assessing its lime requirement and exchangeable hydrogen. The nature of the interaction between an acid soil and a base cannot also be said to have been clearly understood. Also, an adequate explanation of the observation that more base is required to attain a certain pH when the soil is titrated in the presence of a neutral salt than when titrated alone [Crowther and Martin, 1925 ; Hardy and Lewis, 1929 ; Clark and Collins, 1930] has not been so far forthcoming.

In a series of papers being published from this laboratory [Mitra, 1936] the nature of the reactions between colloidal solutions of hydrogen clays and bases both in the presence and absence of neutral salts is being studied in detail. A theoretical formulation of the reactions in some simple systems related to hydrogen clays has been given by Mukherjee, Mitra and Mukherjee [1937]. The present paper being the first part of a series and its main purpose being to examine in the light of this theory the results of comparison of the base binding capacities of hydrogen clays by different methods, the conclusions of Mukherjee, Mitra and Mukherjee [1937] as might be extended to hydrogen clay sols are discussed in some detail below.

The colloidal particles of the sols are surrounded by hydrogen ions existing partly in a free, or, osmotically active condition forming the mobile sheet of an electrical double layer and partly in a secondarily adsorbed state on the surface. The osmotically active hydrogen ions give rise to the observed hydrogen ion activity of the sol ; the remaining hydrogen ions are present in a 'bound', that is, osmotically inactive condition.

In the interaction of the sol with a neutral salt, its cations displace hydrogen ions from the double layer, the amount of displacement depending on the adsorbability of the cations. Where Van der Waal's forces or chemical valence forces do not operate between the oppositely charged ions in the pairs formed by adsorption, the energy of adsorption is determined by their electrical properties, e.g. valency and mobility and the condition of hydration of the ions forming the ion pairs. Consequently, on the addition of different neutral salts having a common anion at the same concentration, the hydrogen ion activity of the sol increases according to the order $Ba^{++} > Ca^{++} > K^{+} > Na^{+}$ of the electrical adsorption of the hydrated cations which is also in agreement with the lyotrope series. This has been called a *regular cation effect*.

In interactions with bases also, the cations of the latter have a marked effect. Apart from the direct neutralization of the free hydrogen ions by hydroxyl ions, the cations of the base displace various amounts of bound hydrogen ions from the double layer which are then neutralized by the OH^{-} ions. The greater the displacement the greater is the amount of acid reacting with the base at a given pH . Titration with different bases thus yields different total acids calculated at a fixed pH . The total acid decreases in the order $Ca(OH)_2 > Ba(OH)_2 > NH_4OH > KOH > NaOH$. In the alkaline region

calcium appears to act more intensely than barium and this has been attributed to what has been called the *irregular cation effect* (*vide* later, p. 347) to distinguish it from the *regular cation effect*.

On titrating a hydrogen clay sol with a given base in the presence of a neutral salt, the cations of the salt present in large numbers, displace hydrogen ions from the double layers and thus bring into a neutralizable condition hydrogen ions present in higher affinity levels in the double layers than those which can be neutralized at the same pH by the base alone.* A greater total acidity is thus obtained on titration in the presence of the salt than in its absence. This explains why in the electrometric titration of soil more base is required to attain a certain pH when the titration is carried out in the presence of a salt than in its absence.

Actually, the total acid is a function of (1) the pH at which it is measured and (2) cation effects. In the absence of a salt, the first additions of the base neutralize mainly the mobile hydrogen ions and the pH rises. The equilibrium between mobile and bound hydrogen ions is disturbed and more hydrogen ions pass from the bound to the mobile condition. These hydrogen ions are then neutralized. The process continues as the pH rises and increasing amounts of the acid are neutralized. Adsorption of the cations of the base as previously explained facilitates this process and here the cation effect comes in. The cation effect finds expression in the different total acidities, measured at the same pH , obtained on titration with different strong bases. In titrations in the presence of a salt, the cation effect is much more marked in view of the large concentration of cations present in the system from the beginning of the titration. The sol+salt mixture contains free hydrogen ions displaced into the intermicellary liquid from the double layers by the cations of the salt and it has been definitely established that the particles in the flocs also contain amounts of hydrogen ions in a reactive condition.** In titrating this mixture, the free hydrogen ions in the supernatant liquid and surrounding the flocs are first neutralized and then as the pH rises more and more hydrogen ions are displaced from the flocs and neutralized. The large number of cations present in the system materially helps this process and the cation effect is emphatically brought out in the larger total acidity, measured at the same pH , obtained on titrating the sol+salt mixture than the sol alone. The hydrogen ions which are brought into a neutralizable condition on the addition of a neutral salt are not all displaced in the intermicellary solution. This is shown by the fact that the titration of the colloid-free extract obtained by continued leaching of the sol with the solution of the salt yields a much smaller total acid than that obtained on titrating the sol+salt mixture *in situ*. This observation is in agreement with that of Ramann already referred to and it shows that lime requirement methods in which the acid displaced in neutral salt extracts of soils is titrated estimate only a part of the total neutralizable acid of soil.

*The cations of the salts also displace Al^{+++} ions whose salts by hydrolysis give rise to some titratable acid in the sol + salt mixture. A systematic study of this point is being carried out in this laboratory by Mr B. Chatterjee.

***Vide* above footnote.

Reference has been made above to the relative effects of Ba^{++} and Ca^{++} ions in the interactions of their salts and bases with hydrogen clays and their dependence on the pH of the system. Added as salts, barium ions have a greater effect than calcium ions in liberating acid from hydrogen clays. Both the hydrogen ion activity and the total acidity of a hydrogen clay sol are increased to a greater extent on the addition of barium chloride than of calcium chloride. The interaction with the salts results in the liberation of acid and takes place in the acid region. The relative effects of the Ba^{++} and Ca^{++} ions in this region follow the electrical adsorption of hydrated ions and thus constitute a *regular cation effect*. But the total acid obtained on titration in the absence of a neutral salt of the cation with calcium hydroxide is, as already observed, usually greater than that obtained on titration with baryta. The Ca^{++} ions thus have a greater effect than the Ba^{++} ions. The slopes of the titration curves point to the same conclusions. An *irregular*, or, *specification effect*, that is, one which does not follow the lyotrope series and is determined by specific forces other than simple electrostatic attraction on the hydrated cations is observed; probably, the cations become dehydrated under these conditions.

In the light of the above, it appears that the lack of agreement between the lime requirement, and the base binding capacities of soils obtained by the different routine methods arises from the fact that different types of cation effect are brought to bear in the different methods depending on the experimental conditions of each as a result of which different amounts of acid are displaced or neutralized even at a given pH , e.g. pH 7.0.

In the present paper, a comparative study has been made of the total acidities of colloidal solutions of hydrogen clays calculated from their electro-metric titration curves as previously obtained in this laboratory* and their base binding capacities obtained by some recognized methods which do not depend on the titration principle. Such comparisons are expected to bring out (1) the significance of the total acidity values calculated from the titration curves in relation to the base binding capacities obtained by the routine methods and (2) the role of cation effects in determining the base binding capacities by such routine methods. The following methods have been used for the comparison:

1. Parker's method [Parker, 1929].
2. Mattson's method [Mattson, 1932].
3. Hissink's back titration method [Hissink, 1925].

It is intended in subsequent papers of this series to extend the work to hydrogen clays obtained from Indian soils other than those used in this work and to compare a larger number of routine methods. It is also intended to extend these studies to soils themselves and to bring the results on hydrogen clays and soils in mutual relation when sufficient experimental material will have accumulated.

*Systematic studies of these curves have been undertaken in a separate series of papers entitled 'On the nature of the reactions responsible for soil acidity'. See, in particular, Part V of this series (R. P. Mitra, this journal, Vol. 6, p. 555, 1936). Parts VI and VII have also been communicated for publication.

Experimental

1. PREPARATION OF COLLOIDAL SOLUTIONS OF HYDROGEN CLAYS

The hydrogen clays were prepared in the manner described in a previous paper [Mitra, 1936] from the clay fractions of the following two Indian soils (surface soils). The clay fractions were separated from the soils using the International soda method.

- (i) Soil from Government Seed Farm, Kalyanpore (U. P.) ; a brown loam.
- (ii) A black cotton soil from Satara Dt., Bombay Presidency ; calcium-saturated, neutral soil.

Hydrogen clay sols H and I respectively were obtained from the above soils.*

2. METHODS OF ESTIMATING TOTAL ACIDITIES AND BASE BINDING CAPACITIES

(a) *Electrometric titration with bases in presence and absence of neutral salts*

The technique followed was as described in a previous paper [Mitra 1936]. Both hydrogen and glass electrodes were used.

(b) *Parker's method*

A known amount of the hydrogen clay was leached with a neutral normal solution of barium acetate. The adsorbed barium estimated as barium sulphate after displacing it by leaching the clay (which was now a barium clay) with a neutral normal solution of ammonium chloride gave the base binding capacity. The adsorbed Ba checked satisfactorily with the adsorbed NH_4 .

(c) *Mattson's method*

To a series of jena glass bottles each containing a definite volume of the hydrogen clay sol was added a neutral salt sufficient to give an approximately normal solution. Increasing amounts of the corresponding base were added to the different bottles. The pH values of the mixtures were then determined using the glass electrode and a titration curve obtained by plotting these pH values against the equilibrium concentrations of the base added. To a second series of bottles containing only the salt solution in the concentration as previously used were also added increasing amounts of the base and on plotting the pH values against the final concentrations of the added base a second curve was obtained. The base binding capacity of the hydrogen clay at any pH was given by the distance (reckoned at this pH) between the two curves parallel to the axis showing the amounts of the base added.

(d) *Hissink's back titration method*

To a given volume of the hydrogen clay sol was added a sufficient amount of the base to make the resulting pH about 11.00. The amount of the base reacting with the hydrogen clay was obtained by conductometrically titrating the excess base with a standard acid. Different bases were used.

RESULTS

The base binding capacities are given in Tables I and II.

*The first soil was obtained through the courtesy of the Superintendent, Government Seed Farm, Cawnpore. The other soil was kindly supplied by the Agricultural Chemist, Bombay.

TABLE I

Base binding capacity of hydrogen clay H obtained by different methods

Method.	Base binding capacity at pH 7.0 in m. e. base per 100 gm. colloid.
A. Electrometric titration in presence and absence of salts.	
Titration with—	
(i) Ba (OH) ₂	32.0
(ii) Ca(OH) ₂	32.8
(iii) NaOH	10.7
(iv) Ba (OH) ₂ in presence of BaCl ₂ (0.83 <i>N</i>)	48.0
(v) Ca (OH) ₂ in presence of CaCl ₂ (0.83 <i>N</i>)	47.0
(vi) NaOH in presence of NaCl (0.83 <i>N</i>)	40.0
(vii) Ba (OH) ₂ in presence of Ba (Ac) ₂ (0.83 <i>N</i>)	51.0
B. Mattson's method using—	
(i) Ba (OH) ₂ and BaCl ₂ (0.83 <i>N</i>)	47.0
(ii) NaOH and NaCl (0.83 <i>N</i>)	39.5
C. Parker's method	
	51.0
D. Hissink's method* using—	
(i) NaOH to give pH 11.1	80.2
(ii) Ba (OH) ₂ to give pH 10.87	85.4
(iii) Ca (OH) ₂ to give pH 10.90	88.2

TABLE II

Base binding capacity of hydrogen clay I obtained by different methods

Method	Base binding capacity at pH 7.0
A. Electrometric titration in presence and absence of salts—	
Titration with—	
(i) Ba (OH) ₂	82.0
(ii) Ca (OH) ₂	97.0
(iii) NaOH	78.0
(iv) Ba (OH) ₂ in presence of <i>N</i> BaCl ₂	110.5
(v) Ca (OH) ₂ in presence of <i>N</i> CaCl ₂	106.0
B. Mattson's method using—	
(i) Ba (OH) ₂ and <i>N</i> BaCl ₂	109.5
(ii) Ca (OH) ₂ and <i>N</i> CaCl ₂	105.0
C. Parker's method	
	110.5

*The base binding capacity estimated by this method does not correspond to pH 7.0. It really conforms to the pH finally attained on adding the alkali to the sol.

The results clearly bring out the variability of the total neutralizable acid as previously discussed. The manner of variations is also as previously indicated. Approximate agreement, however, is found to exist between the base binding capacities (at $pH\ 7.0$) obtained by methods A (iv), A (vii), B(i) and C. The agreement between A (iv) and B (i) is expected as B (i) (Mattson's method using $Ba(OH)_2$ in presence of $BaCl_2$) really amounts to a titration of the sol + salt mixture as carried out in A (iv) with the difference that in B (i) the titration is not continuous. B (i) also requires much larger quantities of the hydrogen clays than A (iv). The cation effects brought to bear in A (iv) and B (i) are the same and consequently, the same amount of acid is neutralized at a given pH , viz., $pH\ 7.0$.

The agreement with Parker's method (method C) which does not depend on the titration principle is interesting as it gives a definite significance to the total acidity values obtained on titrating the sol in the presence of barium chloride [A (iv)] and barium acetate [A (vii)]. An explanation of this agreement may be given in the light of the theoretical considerations already brought forward. When barium chloride or barium acetate is added to the colloidal solution of the hydrogen clay, the H^+ ions present in the double layers are exchanged for Ba^{++} ions. This interchange, however, being a reversible process, all the H^+ ions are not exchanged and at equilibrium, the relative distribution of Ba^{++} and H^+ ions in the double layer is determined by the distribution of these ions in the bulk of the liquid phase. The addition of baryta removes some H^+ ions from the liquid phase whose Ba^{++}/H^+ ratio thus increases. A new equilibrium between the solid and the liquid phases is, therefore, set up which requires a higher Ba^{++}/H^+ ratio in the double layer than what previously obtained. This latter ratio increases as the pH of the system rises, there being a definite relation between the absolute values of the ratios in the bulk of the liquid phase and in the double layer. Thus the amount of Ba absorbed, or, conversely, the amount of H^+ ions displaced from the double layer and neutralized is, as already explained, a function of the pH , the Ba^{++} ion concentration in the liquid phase remaining practically constant (N). At a different Ba^{++} ion concentration, the amount of acid neutralized at the same pH would be different. The smaller total acids of the sols measured at $pH\ 7.0$ obtained on titration with baryta alone than in the presence of barium chloride would be thus explained.

When the sol is leached with a solution of barium acetate as in Parker's method, the Ba^{++} ions of the leaching solution displace H^+ ions from the double layers; these H^+ ions mostly form undissociated acetic acid molecules and are removed from the sphere of action as fast as they are displaced by the Ba^{++} ions. The colloidal particles thus always find themselves in a medium having $pH\ 7.0$ and a normal Ba^{++} ion concentration. The conditions obtaining after the leaching has sufficiently progressed are identical with those reached on titrating the sol + $BaCl_2$ (N) mixture to $pH\ 7.0$ in so far as the Ba^{++}/H^+ ratio in the liquid phase is concerned. Under these conditions, the amount of Ba adsorbed from the barium acetate solution becomes, for reasons stated above, identical with the total acidity of the sol + $BaCl_2$ (N) mixture.*

*The slightly smaller total acid of the sol + $BaCl_2$ mixture compared to the base binding capacity by Parker's method is probably due to the mixture containing $0.83N$ $BaCl_2$ instead of N $BaCl_2$.

Tables I and II show that Hissink's back titration method yields the highest base binding capacities. The results obtained with different bases are different increasing in the order: $\text{NaOH} < \text{Ba}(\text{OH})_2 < \text{Ca}(\text{OH})_2$. The high values obtained by this method are expected as in the highly alkaline regions obtaining under the conditions of the experiment more H^+ ions associated with the colloidal particles enter into the reaction with the base than would be the case in the neutral, or, acid region. The method is, however, open to the objection that a decomposition of the exchange complex may take place under the conditions of titration [Schofield, 1931]. A 'break down' of the absorption complex of a hydrogen clay from a lateritic soil from Bengal has been observed in this laboratory at pH 12.5. A second criticism against Hissink's method is that it measures the soil hydrogen under conditions of alkalinity which do not usually obtain in soil under field conditions.

Summary

The total acidities of colloidal solutions of hydrogen clays calculated from their electrometric titration curves have been compared with their base binding capacities obtained by other methods. The total acid calculated at pH 7.0 from the titration curves is a variable quantity depending on the nature of the base used and on whether the titration is carried out in the presence or absence of a neutral salt using a given base. Titration of the sol+salt mixture yields a much higher total acid (expressed as m. e. base per 100 gm. colloid) than titration of the sol alone. Titration with baryta in presence of *N*-barium chloride, or, *N*-barium acetate yields the highest total acid and it is equal to the base binding capacity of the hydrogen clay obtained by Parker's method. Much higher values of the base binding capacity are obtained by the back titration method of Hissink. The results have been discussed in the light of cation effects which are brought to bear in the different methods of estimation.

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STUDIES ON SOIL TEMPERATURES IN RELATION TO OTHER FACTORS CONTROLLING THE DISPOSAL OF SOLAR RADIATION*

BY

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(With five text-figures)

INTRODUCTION

IN December 1933 the present writer undertook, at the suggestion of Dr L. A. Ramdas, certain investigations on soil temperatures at the Central Agricultural Meteorological Observatory at Poona. These were intended to form part of a scheme of related investigations on the disposal of the radiation received from the sun and the sunlit sky. Apart from routine observations of soil temperatures a number of experiments were performed, mostly during the clear seasons of the last three years, in order to ascertain how far one can modify the thermal conditions in the soil layers near the surface of the ground. A brief account of the results was published in two recent notes [Ramdas and Dravid, 1934, 1936]. A fuller discussion of these results is attempted in the present paper.

A brief description of the city of Poona and its surroundings and of the general climate of the locality may serve as a useful introduction.

DESCRIPTION OF POONA AND ITS SURROUNDINGS

Poona (Lat. $18^{\circ} 30' N.$, Long. $73^{\circ} 53' E.$) is situated at the confluence of the two rivers, Mutha and Mula, near the western margin of the Deccan plateau at a height of 1,830 ft. above the mean sea level. The city is surrounded by numerous low hills.

The general features of the climate of Poona are brought out by Table I which gives the normals of the various meteorological elements for Poona.

The South-west monsoon sets in in June and continues up to the middle of September. The monsoon season is characterised by over-cast skies, frequent drizzling, high south-westerly or westerly winds and small diurnal variation of temperature. There are also occasional thundershowers in this part of India during the pre-monsoon and post-monsoon months.

The climate during the dry season with which we are concerned mainly in the present paper (November-April) is of the ' dry continental type ' essentially controlled by insolation during day and radiative cooling during night.

* This investigation was carried out while the present writer was working as a research student in the Agricultural Meteorology Section (financed by the Imperial Council of Agricultural Research) at the Meteorological Office, Poona. The paper is a revised form of the thesis submitted to the University of Bombay for the M.Sc. degree.

TABLE I *

Month	Mean daily Maximum (°F.)	Mean daily minimum (°F.)	Range (°F.)	Extreme maximum (°F.)	Extreme minimum (°F.)	Relative humidity (per cent)	Vapour pressure in inches of Hg.	Cloud in tenths of sky covered	Rain in inches	Number of rainy days	Wind (in miles per hour)	Direction
January	86.1	54.2	31.9	94.6	42.5	61	.830	1.3	0.06	0.2	4.5	N58W
February	90.6	56.2	34.4	101.9	38.8	54	.820	0.9	0.06	0.1	5.0	N84W
March	97.1	62.8	34.3	109.1	44.8	46	.849	1.0	0.06	0.2	6.1	N86W
April	101.1	65.9	35.2	109.6	50.5	43	.821	1.4	0.57	1.3	7.6	N82W
May	99.7	71.9	27.8	110.0	57.3	56	.867	2.2	1.20	1.8	10.6	N87W
June	89.6	72.6	17.0	108.5	63.0	73	.701	6.5	4.77	7.6	11.2	S87W
July	82.8	71.0	11.8	96.0	66.3	82	.712	8.0	7.01	12.3	11.6	S80W
August	81.7	69.6	12.1	92.1	62.9	84	.696	7.6	3.66	9.0	10.2	S84W
September	84.6	68.6	16.0	96.0	60.9	82	.684	6.4	4.84	7.5	7.9	W
October	89.1	66.5	22.6	100.0	52.3	73	.612	3.6	3.74	5.2	4.8	N61W
November	86.8	59.4	27.4	96.5	48.0	68	.444	1.9	0.98	1.7	4.5	N 23 E
December	84.7	53.9	30.8	95.0	42.5	61	.344	1.5	0.16	0.5	4.3	N 40 E

* Based on data recorded at Yeravda (Poona) from 1878-1920.

Clear skies, feeble air movements, large diurnal range of temperature and of relative humidity and low water vapour content of the air layers near the ground are the characteristics of this season. The Bombay-Deccan, of which Poona is fully representative, is more or less outside the areas directly affected by the north-east monsoon and the western disturbances during the winter months. The clear season extending over six months of the year in the Bombay-Deccan is, therefore, convenient for investigations on soil temperatures under comparatively simple climatic conditions.

THERMAL BALANCE AT THE SOIL SURFACE

The variation of temperature with depth and time in the soil depends upon a number of factors which control the disposal of solar radiation at the earth's surface. These factors are enumerated below :—

1. The duration and intensity of the total radiation from the sun and the sunlit sky received by unit area of a horizontal surface.
2. The colour of the soil surface which determines what fraction of the incident energy is absorbed by the soil surface.
3. The thermal conductivity of the soil which depends upon :—
 - (a) the chemical composition of the soil,
 - (b) the water content, and
 - (c) the pore space or apparent density.
4. The heat transfer from the heated soil surface by convective processes in the air layers near the ground.
5. The radiative exchange in the long wave-length or infra-red region of the spectrum between the soil surface and the atmosphere.
6. Evaporation and condensation of water vapour at the ground surface [Ramdas and Katti, 1934 ; 1936] ; during the clear season at Poona, the day to day variations in the moisture content of the surface soil are small compared to the diurnal variations, the loss by evaporation during the day being recouped more or less by absorption of water vapour from the atmosphere during night.

To determine the heat balance at the earth's surface it is necessary to make a systematic measurement of each of the above factors. The Central Agricultural Meteorological Observatory at Poona, which was started in 1933, has been slowly improving the equipment necessary for a complete scheme of observations

1. Intensity of radiation from the sun and the sunlit sky and the duration of hours of clear sunshine

The measurement of total radiation from the sun (S) and sky (H) is made at the Central Agricultural Meteorological Observatory by using a Moll Solarigraph which consists of a sensitive thermopile and a recording Millivoltmeter (made by Messrs Kipp and Zonen-Delft). The monthly means of the total daily radiation expressed in gramme-calories, the number of days for which records were available and the mean duration of sunshine as recorded by a Campbell-Stokes Sunshine recorder are given in Table II

TABLE II

Mean daily values of S + H (i.e. energy received from the sun and sky) expressed in gramme-calories per sq. cm., duration of sunshine, etc., during different months of the year 1935.

	January	February	March	April	May	June	July	August	September	October	November	December
Mean daily value of S+H	511	644	725	784	775	585	388	439	526	474	600	473
Number of days of observations	31	28	31	30	31	30	11	5	22	23	8	21
Mean daily hrs. of sunshine	8.5	10.4	10.6	11.2	10.9	7.2	3.1	3.7	5.9	7.3	9.5	8.7
Maximum value of S+H	606	715	794	846	855	817	643	507	680	656	622	575
Minimum value of S+H	274	450	661	691	602	131	116	325	302	261	555	318
Mean of S+H on clear days	558	659	734	799	798	759	691	627	600	515
Number of clear days	14	24	23	19	20	5	1	5	8	10
Mean of S+H on overcast days	375	380	319	325	360	261	...	372
Number of overcast days	4	3	5	1	3	1	...	4

for the different months of the year 1935 [Raman, 1935]. In the same table the highest and the lowest values of $S+H$, recorded during each month, are also given. For the sake of comparison the mean values of $S+H$ on clear days alone and on over-cast days are given separately along with the number of occasions of each type at the bottom of the table.

The mean values of $S+H$ on clear days alone during different months give an idea of the intensity of possible radiation in different seasons. April and May are seen to be the two months in which the possible radiation income is greatest and December shows the minimum possible radiation. No records of clear days are available for the months of July and August, but there is no doubt that the intensity of possible radiation in these months would be intermediate between those of June and September.

2. Albedo of the surface of the ground

It is necessary to see what fraction of the incoming solar energy is actually absorbed and converted into heat by the soil surface for a study of the heat economy at the earth's surface. For this purpose it is sufficient to measure the reflection co-efficient or the albedo of the surface for the visible radiation which preponderates in the solar spectrum. These reflection coefficients were determined by using a Moll thermopile with a glass window and a cone and a sensitive galvanometer. The thermopile was directed towards the sunlit surface under experiment and then towards a standard white surface of French chalk also exposed to full sunshine. The deflection in the first case when divided by the deflection in the second case gives the albedo, if we assume that the chalk surface diffuses all the incident radiation. Measurements were made for the surfaces mentioned in Table III.

TABLE III

Kind of surface	Albedo (per cent)
French chalk	100 (assumed)
Charcoal powder	6
Poona black cotton soil	16
Grass covered soil	32
Sakrand soil	41
Belgaum soil	15
Quartz powder	72

It is interesting to note that the surface of Poona soil absorbs 84 per cent of the incident radiation. In fact, soil temperatures of the order of 75°C are occasionally recorded at Poona during the summer. Other soils

referred to in the Table III are typical Indian soils which are not so absorbing as the black cotton soil of the Deccan.

3. *Thermal diffusivity of the soil*

We shall refer in detail later on to the seasonal variation of the thermal diffusivity of the black cotton soil at Poona. It would, however, be appropriate to refer here to the seasonal variation of soil moisture in the different layers of the soil as this is the most important factor which is responsible for the variations in the thermal diffusivity under field conditions. Weekly determinations of soil moisture at depths of 0, 2 in., 4 in., 6 in., 8 in., 12 in. and 18 in. were made from the middle of July 1935, on the bare plot of the Central Agricultural Meteorological Observatory. The data up to the end of November 1936, i.e. over a period of about sixteen months, are shown in Fig. 1 which gives the isopleths of soil moisture. This diagram illustrates the variation of moisture both with depth and with time. The isopleths are drawn at intervals of 5 per cent (moisture content of soil expressed as percentages of dry weight of soil). From the daily rainfall indicated by the length of the vertical lines in the upper portion of the diagram it will be seen that the spells of rain cause the high moisture content lines to approach the surface and that, during the frequent breaks in the monsoon rains at Poona, the moisture content fluctuates rapidly in the first six inches of the soil. After the withdrawal of the monsoon the surface layers of the soil are subjected to more or less unbroken desiccation during the long spell of dry weather extending from the first week of November 1935 to the beginning of June 1936. It is interesting to note that after the initial desiccation the isopleths remain nearly horizontal during the dry season with the 5 per cent line near the surface and the 25 per cent line at a depth of about 1 foot. The diagram brings out quite strikingly the protecting influence of the dry surface soil on the layers below of which the moisture content never goes below 25 per cent.

The fact that the lower layers of the black cotton soil at Poona have a comparatively steady value of moisture content is due to its high water-holding capacity which sets a limit also to the depth down to which percolation can occur with the light rainfall over this tract.

4. *Convective heat transfer from the ground surface*

Some of the thermal energy which accumulates at the surface during the day time is partly conducted into the lower layers of the soil and partly transmitted to the air layers near the ground by convection. (We shall refer to the heat transfer by radiative processes in the next section). The heat transfer from the ground by convection takes place mostly during day time when the surface is warmer than the air above it. Raman [1936] working at Poona has made a simple apparatus for measuring directly the heat carried away from unit area of the ground surface in unit time. A full account of his method will be found in the paper referred to. Fig. 2 curve (C) shows the hourly values of the heat loss by convection in gramme calories per minute on a clear day (23-4-1936). The curve rises quickly after sunrise attaining a maximum between 14 and 15 hours. Later the value decreases rapidly and becomes negligible after sunset.

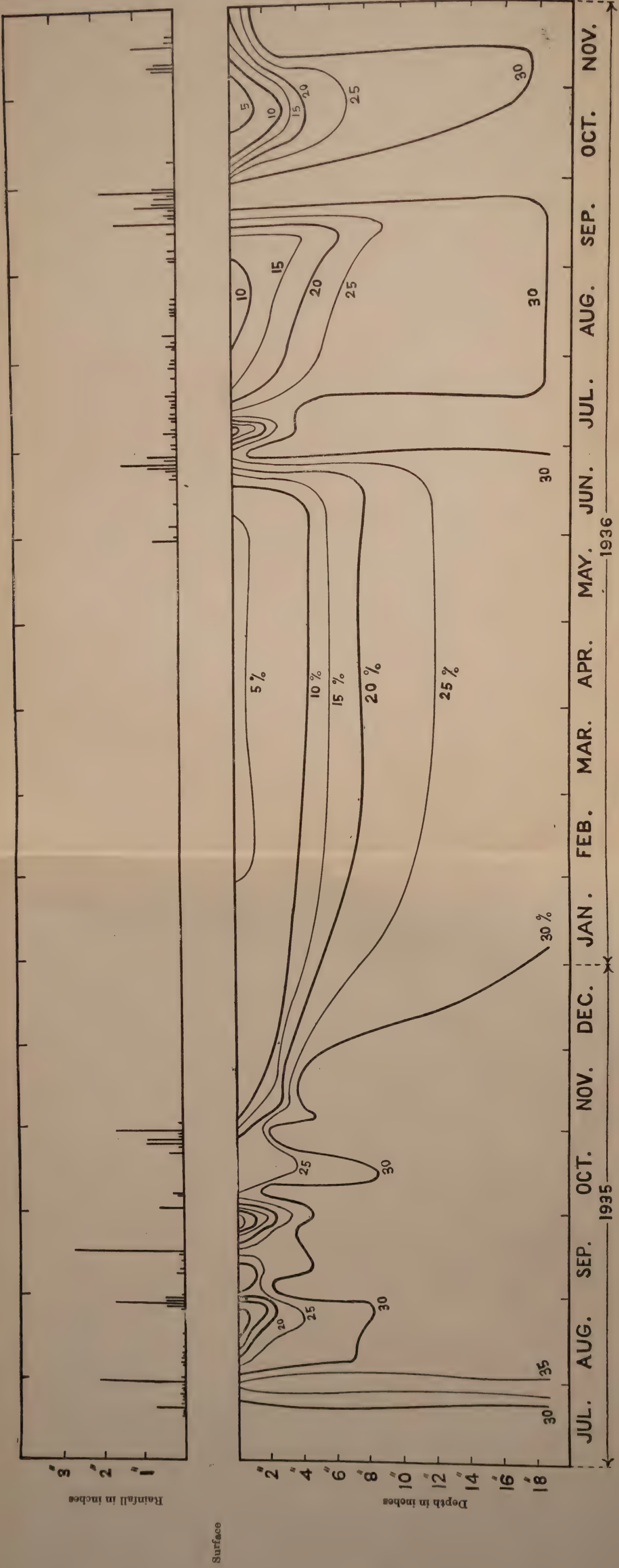


Fig. 1

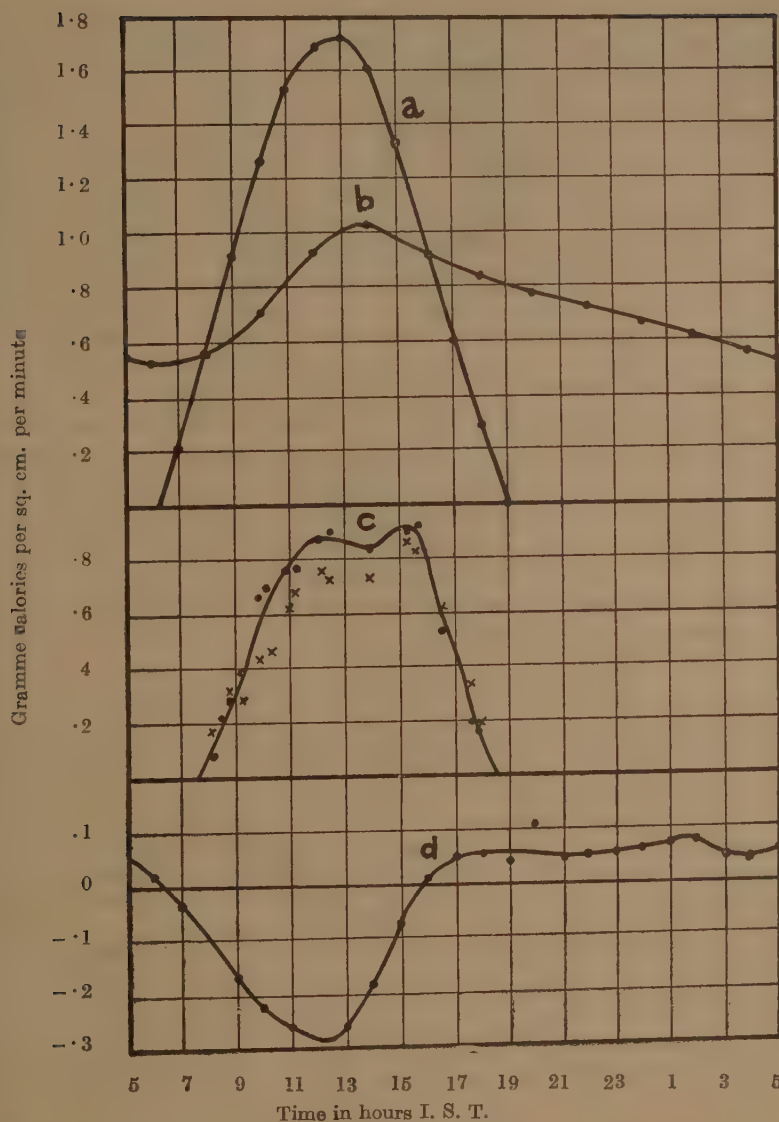


FIG. 2. (23-4-36) (a) Radiation from sun (S) and sunlit sky (H); (b) Temperature radiation going out from the ground surface; (c) Heat loss by convection from the ground surface; (d) Heat transfer by conduction between soil surface and the soil layers below.

5. Radiative exchange in the long-wave-length or infra-red region of the spectrum between the soil surface and the atmosphere

Earth radiation.—During the day time the soil surface is receiving solar radiation in the visible region of the spectrum a part of which it absorbs and converts into thermal energy. The diurnal wave of the surface temperature very closely follows the altitude of the sun and the intensity of the

radiation arriving at the surface from the sun and the sunlit sky. After sunset the surface of the ground begins to cool and the cooling continues until sunrise on the next morning. Bodies at the temperature of the earth's surface emit long wave or heat radiation. The energy radiated has its maximum value at about 10μ , and most of the radiation is confined to the wave-length interval 3μ to 50μ . In this region of the spectrum the surface of the ground emits and absorbs like a black body so that, knowing the surface temperatures T_s (from surface thermograph records or actual hourly observations of surface temperature) we can calculate the total energy E radiated to a hemisphere by unit area of a horizontal surface per second from the relation $E = \sigma T_s^4$ gramme calories per second, where σ is equal to 1.37×10^{-12} and T_s is the surface temperature in degrees absolute. If the atmosphere contained no water vapour all the heat energy radiated by the surface would be lost to space; the atmosphere, however, always contains sufficient water vapour to absorb and re-radiate some of this energy back to the surface of the ground. The heat radiation \bar{S} coming from the night sky as a whole towards unit area of the surface of the ground per minute is measured by means of Angstrom's pyrgeometer. Table IV below gives the mean values of air temperature in degrees absolute, the vapour pressure in mm. of Hg, the black body radiation and \bar{S} the sky radiation, in gramme calories per sq. cm. per minute on clear (cloudless) nights alone during the months of 1934 (excepting July, August and September).

TABLE IV

Mean monthly values of radiation \bar{S} from the night sky, etc., on clear days in 1934

Month	Number of observations	Air temperature degrees absolute	Vapour pressure in mm. of Hg	σT^4 gm. cal/cm. ² minimum	\bar{S} gm. cal/cm. ² minimum
January	7	287	4.7	.556	.386
February	28	295	7.0	.622	.444
March	26	299	9.4	.645	.477
April	22	301	12.5	.671	.490
May	27	302	13.9	.684	.497
June	5	301	17.1	.669	.505
October	3	294	11.3	.607	.463
November	23	290	9.6	.577	.437
December	19	288	7.0	.563	.436

6. *The heat exchange by conduction between the soil surface and the layers soil below*

During day time, when the temperature θ_0 at the surface is higher than θ_1 at unit depth below the surface, the thermal current will flow downwards from the surface. At night when θ_0 becomes lower than θ_1 , the thermal current will flow upwards towards the surface. In general, on a calm and clear day, the heat conveyed downwards and upwards respectively will be roughly of the same order of magnitude, but during transition months when there is an upward or downward trend in the annual variation of temperature there will be some positive or negative carry over to the next day. If θ_0 and θ_1 are the mean temperatures during an hour, the heat conducted into a lower layer through the 1st unit layer with a mean temperature θ_m would be $\lambda (\theta_0 - \theta_1)$ per unit time per sq. cm. and if the unit layer is itself changing in temperature at the rate $\frac{d\theta_m}{dt}$ the accumulation of heat in the unit layer itself will be $C \cdot \frac{d\theta_m}{dt}$ per unit time per sq. cm. where C is the specific heat of the soil. It will be clear, therefore, that the amount of heat conducted from a surface of the soil will be given by $\lambda (\theta_0 - \theta_1) + C \cdot \frac{d\theta_m}{dt}$. Using the appropriate signs for $\theta_0 - \theta_1$, and $\frac{d\theta_m}{dt}$, and knowing the values of θ_0 , θ_1 and θ_m from curves showing the hourly variation of these temperatures it is possible to compute the amount of heat leaving or arriving at unit area of the soil surface per unit time or during hourly intervals.

We are now in a position to consider the thermal balance at the soil surface. We shall consider the conditions on the 23rd April 1936, a clear day during the summer at Poona. Fig. 2, curve (a) gives the march of solar+sky radiation ($S + H$) arriving at the surface. The albedo of the surface for visible radiation may be taken as 15 per cent. The mean value of \bar{S} , the heat radiation from the atmosphere, was 480 gm. cal./cm²/mt. Curve (b) in the same diagram shows the hourly variation of σT^4 or the heat radiation emitted by the soil surface. Curve (c) shows the hourly variation of the heat transferred from the soil surface by convection. Curve (d) shows the heat loss by conduction into the soil layers. In this curve, the portion above the zero-line indicates the gain of heat at the surface by conduction during the night from the lower layers of the soil and the portion below the zero-line indicates the heat lost by conduction into the lower layers from the soil surface during the day hours. The values of the different factors are expressed in Fig. 2 in gramme calories per sq. cm. per minute.

THE SCHEME OF EXPERIMENTS ON SOIL TEMPERATURES

Having discussed the various factors which control the disposal of solar energy at the earth's surface, we may now go on to the subject of soil temperature and its variation with different soils and with different conditions at the surface.

In a note [Ramdas and Dravid, 1934] published in *Current Science* a simple scheme for conducting experiments on soil temperatures has been outlined. As mentioned in the previous section the temperatures attained by different layers of a soil, when its surface is exposed to solar radiation and to the other contemporary meteorological phenomena, will depend to a large extent upon the colour and cover of the surface and the chemical and physical composition of the different layers below the surface.

Johnson and Davies [1927] have measured temperatures at a depth of one centimetre in blocks of tar, macadam, bare earth, sand, rubble, and bare clay 1 metre square and 15 cm. deep. In view of the fact that the samples were 15 cm. deep their results represent the joint effects of the colour and composition of the materials used.

For a preliminary and a comparative study of the behaviour of different typical soils with respect to soil temperatures, the variation due to climatic differences from place to place was eliminated by bringing sufficiently large amounts of the selected soils to one place of observation, viz. the Central Agricultural Meteorological Observatory at Poona.

The experiments were made in distinct stages as follows :—

1. The physical and chemical properties of the soil were kept identical by working with plots of the undisturbed local soil; the plots measured $6\frac{1}{2}$ ft. by $3\frac{1}{2}$ ft. each and similar sets of thermometers were installed at the different depths.

The type of the thermometer used (manufactured by R. Fuess) has a bend near the bulb which makes it easy to fix the bulb at a definite depth, the whole length of the mercury in the bulb lying horizontally at the depth at which the temperature has to be measured. It is also convenient to take the temperature readings from the scale attached to the outer tube surrounding the slender stem, as the stem is inclined to the vertical away from the observer. The thermometer reads correct to one-tenth of a degree Centigrade. The soil thermometers were all compared with a standard thermometer before installation. Corrections, if any, were applied to the recorded observations.

After installing the thermometers at the required depths, comparative observations were taken to verify that the temperatures at corresponding depths were similar. One of the plots (A plot) was kept as a permanent 'control' plot and each of the remaining plots covered with thin layers of substances like chalk, charcoal powder and typical soils from different parts of India. The simultaneous observations were then continued in order to record the influence of the 'cover' on the temperatures of the soil layers below.

Along with these experiments it was also arranged to measure the effects of surface wetting and of a cover of vegetation on the soil temperatures.

2. Having ascertained the effect of cover, the effect of varying both the physical and chemical composition of the soil was studied by using blocks of different soils measuring $6\frac{1}{2}$ ft. in length, $3\frac{1}{2}$ ft. in breadth and 1 ft. in depth. The soil blocks were kept with their natural surfaces exposed in the first part of the experiment and, after comparative observations had proceeded for a sufficiently long time, all except the local 'control' plot were covered with

a thin layer of the local black cotton soil so as to eliminate the influence of the surface colour and retain only the variations due to the interior of the blocks of different soils.

EXPERIMENTS ON THE EFFECT OF SOIL COVER

The experiments carried out in order to study the effect of various soil covers are taken up for discussion in the order in which they were carried out.

Experiment 1: Effect of a thin cover of French chalk on soil temperatures

The soil thermometers were installed in the standard plot A and in the experimental plot B, at depths of 0 cm., 5 cm., 10 cm., 15 cm., 20 cm. and 30 cm.

Two hourly observations were taken of the temperatures shown by the two sets of thermometers at the different depths, after the conditions in the soil layers in the two plots had become normal. It was thus ascertained that the temperatures of the different layers of the soil in the two plots were similar. After the observations had been continued for a sufficient number of days, the plot B was covered with a thin layer (about 1 mm.) of French chalk powder (white in colour) uniformly all over the surface, so that there was no patch of the local black cotton soil left bare and exposed to the sun. The plot A was left untreated and used as a permanent 'control' plot.

The two hourly observations were continued as before. The white surface of the B plot reflected most of the solar radiation and absorbed very little solar radiation whereas the black surface of the standard A plot absorbed about 85 per cent of the energy which would be diffused by the white surface of chalk. Naturally, the temperatures at the different depths in the B plot were considerably lowered as compared to the corresponding temperatures of the A plot. Of course, these changes of temperature took place immediately at the surface but reached their full values only after two days at 10 cm. depth, and four days at 50 cm. depth.

For a convenient discussion of these data, the observations were grouped in weeks according to the scheme given by Sir Napier Shaw, in his paper *The Book of the Grower's year*.

Table V gives the weekly average temperatures of the A and B plots at the depths of 0, 5, 10, 15, 20 and 30 cm. and at the times 0600 hrs., 0800 hrs., 1000 hrs., 1400 hrs., 1600 hrs. and 1800 hrs.

During week No. 1 (25th to 31st of December 1933), both the A and B plots were in their natural untreated condition. It can be seen that the temperatures at different depths are very similar in both the plots at the different epochs during this week. At 1700 hrs. on the 31st of December 1933, plot B was covered with a very thin layer of French chalk powder. The lowering effect in the temperature of plot B was observed during the next three weeks when the chalk cover was retained, viz.—

Week No. 2, January 1 to January 7, 1934,

Week No. 3, January 8 to January 14, and

Week No. 4, January 15 to January 20.

TABLE V

Effect of a thin cover of chalk powder on soil temperatures in °C. (Plot A: Control plot; Plot B: Experimental plot to which the cover of chalk powder was applied at 1700 hrs. on 31st December 1933; the cover was removed at 1700 hrs. on 20th January 1934)

Depth	No. of week	0600 hrs.		0800 hrs.		1000 hrs.		1400 hrs.		1600 hrs.		1800 hrs.	
		A	B	A	B	A	B	A	B	A	B	A	B
0 cm.	1	11.0	11.2	17.3	17.4	30.4	30.3	44.9	44.9	34.4	33.8	25.6	25.5
	2	14.7	14.3	19.5	16.8	30.5	23.3	42.1	31.3	35.3	28.1	27.2	23.8
	3	12.1	10.9	17.6	13.7	33.5	21.9	50.1	31.9	41.9	29.3	28.9	23.4
	4	9.8	8.4	14.8	12.2	32.2	23.9	50.6	36.7	41.9	32.1	29.3	24.1
	5	11.9	11.6	16.4	16.9	33.4	33.4	49.6	48.8	42.0	40.9	28.6	27.6
	6	12.7	12.6	18.4	18.5	31.5	31.0	46.5	45.3	42.2	41.4	23.6	23.3
5 cm.	1	17.3	17.1	16.9	16.7	24.6	24.6	30.1	30.3	29.1	29.3	28.1	28.1
	2	19.5	18.1	19.0	17.5	22.7	20.3	28.7	24.0	28.8	24.4	27.1	23.6
	3	18.9	16.4	18.3	15.9	22.0	18.2	31.3	24.1	31.7	24.7	29.3	23.7
	4	17.2	14.7	16.5	14.1	20.7	16.6	31.1	23.7	31.9	24.6	29.5	23.3
	5	18.0	17.3	17.7	16.9	20.3	19.7	31.8	31.3	31.9	31.1	28.8	27.7
	6	19.2	18.7	18.7	18.4	22.1	22.2	31.7	31.9	31.9	32.1	29.6	29.1
10 cm.	1	20.6	20.3	19.6	19.3	23.0	22.9	24.9	24.9	26.1	26.0	28.0	23.0
	2	21.7	20.1	20.9	19.3	21.9	20.4	24.8	21.8	26.5	23.1	26.5	23.3
	3	21.8	19.3	20.8	18.4	21.3	18.6	26.1	21.4	28.2	23.9	28.3	23.1
	4	20.5	17.5	19.5	16.8	19.8	16.8	24.9	20.1	27.2	22.0	27.4	22.1
	5	21.0	20.1	20.3	19.2	18.9	17.8	25.0	24.6	26.5	26.3	26.2	25.8
	6	22.2	21.9	21.2	20.9	21.6	21.3	26.1	26.1	27.1	27.1	27.5	27.4
15 cm.	1	22.6	22.5	21.8	21.7	23.7	23.7	23.0	22.9	25.9	25.9	26.5	26.5
	2	23.1	21.7	22.5	21.1	22.7	21.5	23.0	21.5	24.7	23.4	25.3	22.8
	3	23.6	21.0	22.9	20.5	22.6	20.3	24.0	20.9	25.7	21.9	26.4	22.4
	4	22.5	19.6	21.7	19.1	21.3	18.7	22.7	19.5	24.5	20.7	23.2	21.1
	5	22.4	21.9	22.0	21.3	20.3	19.4	23.0	22.3	24.1	23.5	24.2	23.7
	6	23.9	23.7	23.1	22.9	23.0	22.8	24.2	24.1	24.8	24.9	23.6	25.8
20 cm.	1	23.6	23.4	23.1	22.9	24.4	24.3	22.8	22.9	24.5	24.8	24.9	25.4
	2	23.7	22.4	23.4	22.0	23.5	22.4	23.1	21.9	23.6	23.4	24.1	22.7
	3	24.3	22.0	23.0	21.6	23.8	21.4	23.7	21.4	24.2	21.8	24.1	22.1
	4	23.4	20.6	22.9	20.2	22.6	19.9	22.5	20.0	23.1	20.6	23.5	20.9
	5	23.2	22.6	22.9	22.1	21.7	20.6	22.6	22.0	23.8	22.5	22.5	22.5
	6	24.5	24.5	24.1	23.8	24.1	23.9	23.8	23.9	23.8	21.1	21.2	24.8
30 cm.	1	24.4	24.4	24.3	24.2	25.2	25.1	24.1	23.9	24.8	24.7	24.9	24.8
	2	24.3	23.2	24.2	23.2	24.3	23.3	24.1	23.1	24.1	23.0	24.2	23.0
	3	25.0	23.0	24.9	23.0	24.9	23.0	24.7	22.8	24.5	22.7	24.5	22.7
	4	24.1	21.8	23.9	21.7	23.9	21.7	23.7	21.6	23.6	21.5	23.6	21.4
	5	23.7	23.1	23.7	23.1	23.5	22.2	23.4	22.7	23.3	22.5	22.7	22.0
	6	25.0	25.0	24.8	24.8	25.1	25.0	24.5	24.5	24.3	24.3	24.3	24.3

The differences in temperatures are relatively small in the morning but in the afternoon they show up very conspicuously. Thus the effect of the French chalk cover is to lower the temperatures at 6 a.m. by 1.2°C , 2.5°C , 2.5°C , 2.6°C , 2.3°C and 2.0°C at depths of 0, 5, 10, 15, 20 and 30 cms. respectively. The temperatures of the covered plot are lowered by 18.2°C , 7.2°C , 4.7°C , 3.1°C , 2.3°C and 1.9°C respectively at 2 p.m. These lowerings in temperatures have been noted from the average temperatures during week No. 3, i.e. January 8 to 14, 1934, the second week after the cover of French chalk was applied. Similarly the lowerings in the temperatures at the various depths due to the white chalk cover may be noted at the different times of observation.

Figs. 3 (a) and 3 (b) are isopleths of the weekly mean temperatures at 1400 hrs. (afternoon) in the control and chalk-covered plots respectively. The abscissae refer to the successive weeks and the ordinates refer to the depths below surface. The plots were similar during the first week. The very conspicuous lowering of the soil temperatures during the second, third and fourth weeks in the chalk covered plot is shown by the rapid approach of the isotherms towards the surface.

The chalk powder was removed from the surface of the B plot at 1700 hrs. on the 20th January 1934 after having remained for three successive weeks. It is interesting to note the gradual return of the isotherms in the treated plot to their normal values. It took more than a week after the removal of the chalk for the temperatures to equalize in the two plots. During the first week after the removal of the cover (January 21 to 28) the temperatures are still seen to be differing in value. But, in the next week (January 29 to February 1) the temperatures are again more or less similar in the two plots.

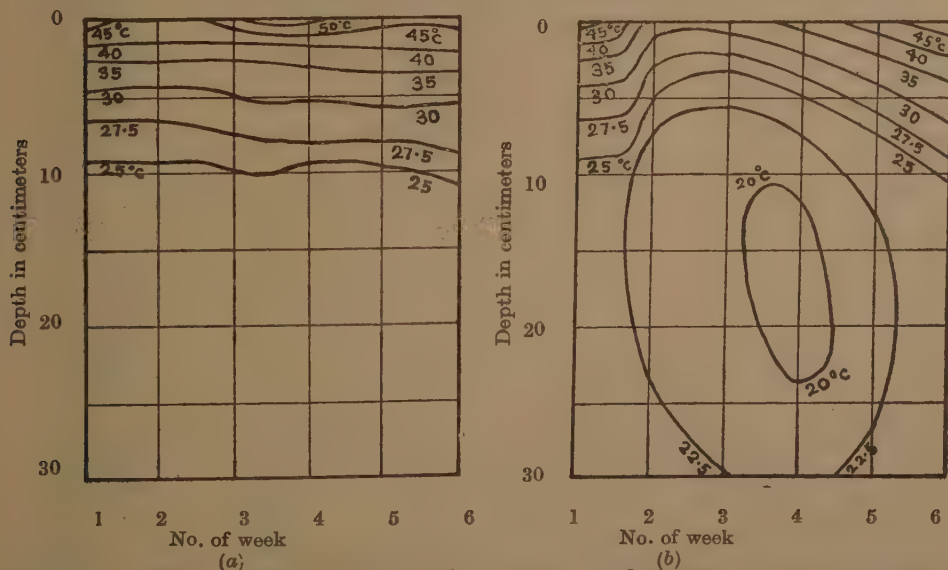


FIG. 3. Effect of a thin layer of chalk powder on weekly mean soil temperatures at 1400 hrs. I. S. T. (25-12-33 to 1-2-34). (a) Control; (b) Chalk put on at the beginning of the 2nd week. Chalk removed at 1700 hrs. of 20-1-34 (end of 4th week).

Experiment 2: Effect of a thin cover of charcoal powder on soil temperatures

After the conditions in the B plot had become normal and the temperatures in both the plots had become similar, a thin cover of charcoal powder was laid on the surface of the B plot uniformly at sunset (1818 hrs.) on the 1st of February, 1934.

Table VI gives the weekly average temperatures at the maximum and minimum epochs, i.e. 0600 hrs. and 1400 hrs. at the various depths in the two plots. The dates corresponding to the different periods are as follows:—

No. of period	Dates
1	29-1-34 to 1-2-34
2	2-2-34 to 11-2-34
3	12-2-34 to 18-2-34
4	19-2-34 to 25-2-34
5*	5-3-34 to 11-3-34
6	12-3-34 to 18-3-34
7	19-3-34 to 25-3-34

* Observations were not recorded during the period 26th February to 4th March 1934.

Evidently, the black charcoal cover should absorb more solar radiation and consequently raise the temperatures in B plot. Since, however, the local Poona soil is also black, there is not much difference in the 'albedo' or colour effect of the soil and the cover (charcoal powder) used. Naturally, such large differences of temperature as were obtained in the first experiment with chalk cover cannot be expected in this experiment. All the same, a distinct rise in temperatures in plot B can be noticed.

TABLE VI

Effect of a thin cover of charcoal powder on soil temperatures in °C (Plot A: Control plot; Plot B: Experimental plot to which the cover of charcoal powder was applied at 1818 hrs. sunset, on 1st February 1934; the cover was removed at 1700 hrs. on 18th March 1934)

No. of week	Depth	0600 hrs.		1400 hrs.	
		A	B	A	B
1	0 cm.	12.7	12.6	46.5	45.3
2		11.9	12.4	53.8	54.7
3		14.1	14.5	55.7	57.4
4		12.3	12.9	56.0	57.9
5		13.6	14.1	56.6	59.1
6		14.5	15.1	57.1	59.0
7		19.6	19.5	58.4	58.3
1	5 cm.	19.2	18.7	31.7	31.9
2		18.9	18.9	35.3	36.3
3		21.2	21.4	38.2	39.5
4		20.5	20.6	37.7	39.5
5		20.9	21.5	38.5	41.2
6		21.6	22.2	38.7	41.4
7		25.6	25.4	42.3	42.6

TABLE VI—*contd.*

No. of week	Depth	0600 hrs.		1400 hrs.	
		A	B	A	B
1 } 2 } 3 } 4 } 5 } 6 } 7 }	10 cm.	22.2 22.3 24.5 24.5 25.0 25.5 28.2	21.9 22.6 25.0 25.1 25.7 26.4 28.6	26.1 27.3 30.4 30.2 31.5 31.8 34.8	26.1 27.6 30.5 30.4 31.7 32.1 34.5
1 } 2 } 3 } 4 } 5 } 6 } 7 }	15 cm.	23.9 24.1 26.2 26.6 27.1 27.5 29.8	23.7 24.4 26.7 27.0 27.7 28.3 29.9	24.2 24.7 27.1 27.2 27.7 28.4 30.8	24.1 25.2 27.8 27.9 28.7 29.3 31.2
1 } 2 } 3 } 4 } 5 } 6 } 7 }	20 cm.	24.5 24.7 26.6 27.1 27.6 28.1 29.9	24.5 25.0 27.2 27.7 28.3 28.9 30.3	23.8 24.0 26.0 26.5 27.1 27.6 29.6	23.9 24.6 27.0 27.3 28.0 28.6 30.3
1 } 2 } 3 } 4 } 5 } 6 } 7 }	30 cm.	25.0 25.0 26.7 27.5 27.9 28.4 30.0	25.0 25.4 27.3 28.1 28.7 29.1 30.3	24.5 24.7 26.4 27.2 27.7 28.2 29.8	24.5 25.1 27.0 27.8 28.3 28.8 30.0

Thus, the effect of the cover of charcoal powder is to raise the temperatures in plot B at 6 a.m. by 0.5°C, 0.6°C, 0.7°C, 0.6°C, 0.7°C and 0.8°C and at 2 p.m. by 2.5°C, 2.7°C, 0.2°C, 1.0°C, 0.9°C and 0.6°C respectively at the depths of 0, 5, 10, 15, 20 and 30 cm. These differences are taken from the average temperatures during the week March 5 to 11, 1934, i.e. the fifth period after the charcoal cover was applied. The cover was carefully removed at 1700 hrs. on the 18th of March 1934. The average temperatures during

the week after the removal of the cover are also given in Table VI against period No. 7. These are more or less similar.

Experiment 3 : Effect of wetting the soil surface with water equivalent in amount to 1/4 in. of rain

After the temperatures in plots A and B had attained normal values plot B was wetted with $\frac{1}{4}$ in. rain equivalent of water sprinkled uniformly all over the surface at sunrise (0625 hrs.) on the 14th of April 1934.

Table VII gives the observations recorded. After two observations at 0600 and 0625 hrs., the plot was watered ; and then up to 8 a.m. observations were taken at intervals of 15 or 20 minutes ; from 8 a.m. to 10 a.m. half-hourly observations were taken and then at 1400, 1500, 1600, 1700 and 1800 hrs. on the 14th of April. From the 15th to 20th April the observations were recorded only at 0600, 0800, 1000, 1400, 1600 and 1800 hrs.

The general effect of the watering is to lower the temperatures in the B plot considerably.

At the surface, we can clearly see how the temperature of the control plot goes on rising after sunrise at the usual rapid rate, while that of the watered plot lags more and more. Whereas at 0640 hrs. the surface temperature of plot B (21.5°C) is slightly higher than that of plot A (21.1°C), at 0700 hrs. the temperature of A (22.4°C) is higher than that of B (21.2°C). At 1000 hrs., we find the surface temperature of control plot (A) higher by 15.8°C and at 1400 hrs. by 14.6°C than the corresponding temperature of the watered plot (B). At 1600 hrs., in the evening, the difference is only 5.8°C . The slower rate of rise and fall of temperatures in plot B is partly due to the increase of the specific heat of the soil due to the presence of water and partly due to the heat utilized for the evaporation which is taking place at the wet surface.

On the 15th April, at the maximum temperature epoch, the surface temperature of plot B is 3.2°C lower than that of plot A, while on the 20th April the difference is only 0.8°C which is almost negligible. Thus after about a week, the lowering effect produced in the surface temperature of the plot B by watering, equivalent to $\frac{1}{4}$ in. of rain, is no longer significant.

The maximum difference between the temperature at 2 cm. depth in the two plots is noticed at 1400 hrs., viz. 11.7°C on the day of watering. On the next day at the same hour, the difference is 5.5°C while on the 20th April it is only 1.5°C .

At the depth of 5 cm. the difference in the temperatures of the two plots is 4.8°C at 1400 hrs. and 5.0°C at 1500 hrs. and 1600 hrs. on the date of wetting. On the next day, the difference at 1400 hrs. is 3.0°C .

The lowerings in the temperatures at 2 p.m. at the depths of 10 cm., 15 cm., and 20 cm. in plot B are 2.6°C , 1.0°C and 0.0°C on the 14th April and 2.5°C , 1.4°C and 0.4°C on the 15th April. Up to 6 p.m. on the day of watering the temperature at the depth of 20 cm. has not been in the least affected in the B plot, but at 6 a.m. the next day we see a fall of 1°C in the temperature. We thus observe that the effect of cooling travels very slowly ; it takes more than 12 hrs. to reach a depth of 20 cm.

TABLE VII

Effect of wetting the surface of the soil on soil temperatures in °C. (Plot A: Control plot; Plot B: Experimental plot of which the surface was wetted with a quantity of water equivalent to $\frac{1}{4}$ in. rain at 0625 hrs. sunrise on 14th April 1934)

Date	Time	0 cm.		2 cm.		5 cm.		10 cm.		15 cm.		20 cm.	
		A	B	A	B	A	B	A	B	A	B	A	B
14th April 1934	0600	19.2	19.8	22.5	23.5	26.2	26.5	30.0	30.6	32.2	32.4	32.9	33.0
	0625	19.8	20.4	23.0	23.8	26.2	26.5	29.8	30.4	32.0	32.2	32.8	33.0
	0640	21.0	21.5	23.2	24.6	26.2	26.4	29.7	30.2	32.0	32.2	32.8	32.8
	0700	22.4	21.2	24.0	24.8	26.2	26.5	29.6	30.2	31.9	32.1	32.8	32.8
	0715	24.5	22.2	25.0	25.0	26.5	26.8	29.5	30.0	31.8	32.0	32.8	32.7
	0730	26.4	23.3	26.4	25.5	26.9	27.1	29.5	30.0	31.8	32.0	32.8	32.6
	0745	27.6	23.3	27.5	25.7	27.6	27.4	29.5	30.0	31.7	31.8	32.7	32.6
	0800	29.6	23.3	28.5	25.8	28.3	27.7	29.5	30.0	31.6	31.8	32.6	32.5
	0830	33.5	25.6	30.4	26.5	29.5	28.0	29.8	30.0	31.6	31.7	32.6	32.5
	0900	40.0	29.2	34.2	28.5	31.0	28.8	30.6	30.3	31.5	31.6	32.5	32.4
	0930	44.8	32.2	38.0	31.0	32.4	30.0	31.2	30.4	31.5	31.6	32.4	32.3
	1000	49.0	33.2	40.8	32.8	34.3	31.3	37.4	34.8	33.8	32.8	32.5	32.5
	1400	68.8	54.2	56.0	44.3	46.1	41.3	39.0	36.1	34.8	33.5	32.8	32.8
	1500	67.0	54.1	56.3	45.0	47.4	42.4	39.9	36.9	35.4	34.0	33.1	33.1
15th April 1934	1600	61.9	52.2	55.0	44.9	47.6	42.6	39.9	36.9	35.4	34.0	33.4	33.4
	1700	53.0	46.0	49.5	41.9	46.2	41.5	40.2	37.2	36.0	34.4	33.8	33.8
	1800	42.9	37.1	45.5	38.0	43.6	38.8	39.8	37.0	36.6	34.8	33.8	33.8
	0900	20.0	18.8	24.2	24.0	28.0	26.8	31.0	30.0	32.6	31.6	33.2	32.2
	0930	30.0	28.8	29.2	27.0	28.6	27.2	30.3	29.5	32.2	31.2	33.0	32.0
	1000	52.1	49.6	42.8	38.5	37.0	34.6	32.4	31.0	32.1	31.2	32.8	31.7
	1400	64.0	60.8	55.5	50.0	46.8	43.8	39.0	36.5	34.8	33.4	33.0	32.6
	1600	59.6	56.6	52.3	47.5	46.0	43.2	39.8	37.4	35.6	34.1	33.4	33.0
	1800	40.7	38.8	42.8	38.8	42.1	39.8	39.7	37.3	36.8	35.8	34.0	33.6

TABLE VII—*contd.*

Date	Time	0 cm.		2 cm.		5 cm.		10 cm.		15 cm.		20 cm.	
		A	B	A	B	A	B	A	B	A	B	A	B
16th April 1934	0600	22.2	21.6	26.0	26.0	29.4	28.8	32.4	31.2	33.6	32.4	33.6	32.8
	0800	29.2	28.8	29.0	28.0	29.4	28.6	31.6	30.8	33.0	32.0	33.2	32.6
	1000	44.5	44.8	40.0	37.0	35.2	33.7	31.6	31.3	32.8	31.9	33.2	32.4
	1400	68.4	64.8	56.2	51.0	46.8	44.2	38.8	36.7	34.8	33.5	33.2	32.0
	1700	47.6	46.5	47.8	44.8	44.8	43.2	40.4	38.8	36.8	35.6	34.0	33.2
	1800	40.0	39.2	43.2	41.2	42.6	41.4	39.6	38.3	37.0	35.7	34.4	34.4
17th April 1934	0600	22.7	22.3	27.0	27.0	30.1	29.2	32.6	31.6	33.6	32.8	33.7	33.0
	0800	29.2	29.0	28.0	28.0	29.8	28.8	31.6	31.0	32.9	32.2	33.4	32.8
	1000	40.4	45.0	40.0	37.1	35.6	34.0	32.4	31.5	32.7	32.1	33.4	32.6
	1400	65.8	62.0	52.5	49.5	44.6	43.2	38.4	36.4	34.8	33.8	33.2	32.6
	1600	55.8	54.8	49.2	47.0	43.3	43.8	39.8	38.2	36.0	34.9	33.8	33.2
	1800	41.8	39.8	42.8	41.2	41.5	41.0	39.0	38.0	36.6	35.4	34.2	34.4
18th April 1934	0600	21.6	21.8	25.4	25.9	28.4	28.4	31.4	31.1	33.2	32.6	33.6	33.2
	0800	30.4	30.0	29.0	28.6	29.0	28.6	30.8	30.2	32.5	31.8	33.2	32.6
	1000	47.0	46.0	39.0	37.5	34.5	33.4	32.4	31.4	32.4	31.8	33.0	32.4
	1400	60.2	58.0	51.0	47.0	43.8	42.0	37.4	36.0	34.2	33.4	33.0	33.0
	1600	51.8	49.6	47.7	44.5	43.0	42.2	38.8	37.8	35.4	34.5	33.6	33.6
	1800	37.0	35.1	41.8	38.8	40.7	40.0	38.0	37.5	36.0	35.0	34.0	34.2
19th April 1934	0600	18.6	18.9	23.2	24.0	27.0	27.8	30.6	30.2	32.5	31.9	33.2	32.6
	0800	35.8	34.8	31.5	30.0	29.2	28.7	29.8	29.4	31.6	31.0	32.6	32.0
	1000	49.6	48.0	39.7	37.0	34.1	33.0	31.8	30.5	31.6	31.1	32.5	31.8
	1400	61.2	59.2	51.2	48.0	43.2	42.9	37.0	35.7	33.2	32.2	32.4	32.4
	1600	52.2	50.8	46.8	43.7	42.8	41.1	36.6	35.4	33.2	32.3	33.3	33.3
	1800	39.5	37.9	40.1	38.2	40.0	38.9	37.8	37.0	35.7	34.9	33.5	33.8
20th April 1934	0600	21.2	21.6	24.8	25.2	27.6	28.0	30.8	30.8	32.6	32.0	33.0	32.6
	0800	26.2	26.4	27.0	27.5	27.6	27.5	30.0	30.2	32.0	31.6	33.0	32.2
	1000	46.9	45.4	37.8	35.8	33.6	32.8	31.3	30.6	31.1	31.3	32.5	31.9
	1400	56.8	56.0	47.0	45.5	41.2	42.0	37.4	36.8	34.5	33.8	32.8	33.0
	1600	51.8	51.5	45.0	44.3	40.8	41.6	38.1	37.8	35.7	34.2	33.4	33.4
	1800	39.6	38.2	40.4	39.3	39.5	39.2	37.0	37.1	35.7	34.7	33.5	33.9

Experiment 4: Effect of wetting the surface of the soil with water equivalent in amount to $\frac{1}{2}$ in. of rain

After the soil temperatures in the two plots A (control) and B (experimental) had become more or less similar for a number of days the surface of the plot B was wetted uniformly with water equivalent to $\frac{1}{2}$ in. of rain. From Table VIII it will be seen that the effect of wetting plot B on the morning of the 3rd May (6 a.m.) has been to depress the afternoon soil temperatures in that plot by 11.1°C, 14.8°C, 10.7°C, 4.4°C, 3.1°C, 2.0°C, 0.4°C and 0.2°C at depths of 0, 0.5, 2, 5, 10, 15, 20 and 30 cm. respectively. The recovery from the effect of wetting is perceptible even on the next day; but it is only after the 6th May that the temperatures in the two plots become more or less equal.

TABLE VIII

Soil temperatures in °C in two plots A (control) and B (experimental) during the period 3rd to 6th May, 1934, showing the effect of wetting the surface of the plot B on the morning of 3rd May 1934

Morning 6 a.m.

Dates	3-5-34		4-5-34		5-5-34		6-5-34	
Depth cm.	A	B	A	B	A	B	A	B
0	22.8	23.8	21.3	20.2	19.2	18.5	16.6	17.1
0.5	24.0	24.7	22.4	20.6	20.5	19.6	18.5	18.0
2	27.0	27.0	25.2	23.0	24.0	23.0	22.0	21.5
5	29.1	29.4	28.0	25.9	27.3	26.0	25.8	25.2
10	31.6	32.0	30.4	28.7	30.6	29.8	29.6	29.5
15	33.2	33.2	32.7	30.2	32.5	31.3	32.1	31.0
20	33.6	33.8	33.6	32.0	33.4	32.0	33.1	32.1
30	33.8	33.8	33.9	32.9	33.8	32.8	33.6	32.6

Afternoon 2 p.m.

Dates	3-5-34		4-5-34		5-5-34		6-5-34	
	A	B	A	B	A	B	A	B
0	66.4	55.3	63.3	59.8	62.1	60.0	64.5	63.5
0.5	61.0	46.2	57.8	53.4	56.5	54.1	59.8	57.8
2	50.8	40.1	48.0	42.1	46.4	43.3	49.2	46.5
5	41.6	37.2	39.8	36.1	37.8	37.5	39.8	39.6
10	36.9	33.8	36.0	33.3	35.3	33.2	35.6	33.6
15	34.4	32.4	33.8	31.7	33.5	31.6	33.0	31.5
20	33.3	32.9	33.2	32.0	33.0	32.2	32.4	31.9
30	33.5	33.3	33.6	32.6	33.4	32.5	33.1	32.4

Figs. 4 (a) and 4 (b) are the isopleths of daily temperatures at 1400 hrs. in the control and surface-wetted plots respectively. The wetting was done at 6 a.m. on the third day. The sudden cooling communicated to the various soil layers is shown by the rapid approach of the isotherms towards the surface on the third day. The recovery from the effects of wetting was gradual and the temperatures had not yet equalized even on the 6th day.

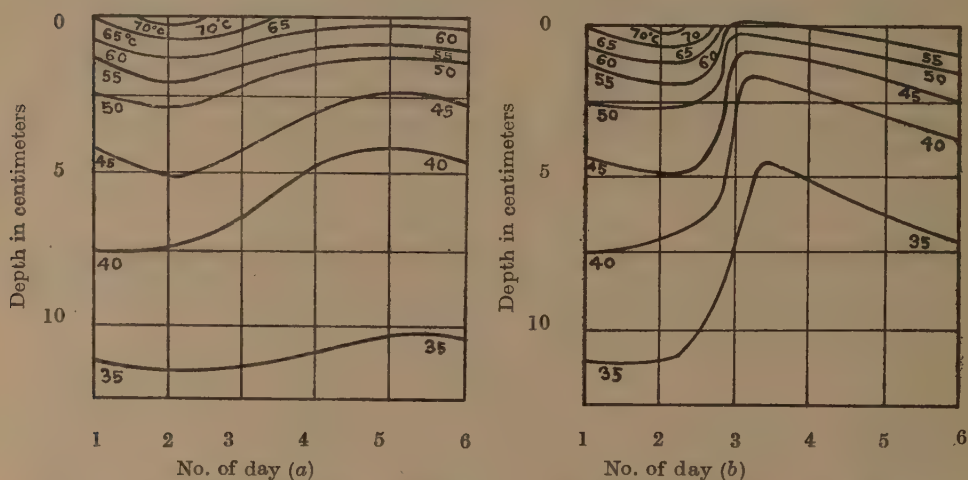


FIG. 4. Effect of watering the surface on daily soil temperature at 1400 hrs. I. S. T. (1-5-34 to 6-5-34). (a) Control ; (b) Surface moistened on 3-5-34.

Experiment 5 : Effect of a cover of vegetation on soil temperatures

After making a survey of the effects of changing the surface cover on soil temperatures as outlined in the preceding paragraphs, an attempt was made to study the effect of a cover of vegetation on the temperatures of the soil. The three plots A, B and C were used for this experiment. Plot A was used as 'control'. After comparative observations under similar conditions had been made for some days, plot C was sown with 'aleev' on September, 4th, 1935. Plot B was kept bare but received periodical watering to the same extent as plot C. The height of the 'aleev' crop was 24 cm. on the 11th of October, 28 cm. on the 19th of October, and 30 cm. on 4th of November. It was trimmed to 20 cm. on 17th of November. Table IX shows the weekly average temperatures at 0600 hrs. and 1400 hrs. at depths of 0, 2, 5, 10, 15 and 20 cm. for the following weeks :—

Week No.	Dates
1	22nd to 28th August 1935
2	20th to 26th November 1935
3	27th November to 3rd December 1935

TABLE IX

Morning and afternoon average soil temperatures during three weeks in the three plots A, B, C, at different depths showing the effect of a plant cover on the plot C, the plot B being given the same amount of water as the plot C and the plot A being left untreated as the standard control plot. Week 1 before treatment ; weeks 2 and 3 after treatment

Depth in cm.	No. of week	Morning 0600 hrs.			Afternoon 1400 hrs.		
		Soil temperatures °C			Soil temperatures °C		
		Control plot	Plot receiving equal amount of watering as plot C	Plant covered plot	Control plot	Plot receiving equal amount of watering as plot C	Plant covered plot
		A	B	C	A	B	C
0 cm.	1	21.2	21.2	21.2	40.8	40.8	40.8
	2	9.1	8.8	10.6	54.3	37.8	27.8
	3	10.5	9.7	13.9	54.7	38.8	24.1
2 cm.	1	23.7	23.7	23.7	33.9	33.9	33.9
	2	16.2	12.3	14.9	37.3	28.7	26.7
	3	17.2	13.4	15.9	39.1	30.8	26.0
5 cm.	1	24.6	24.6	24.6	31.7	31.7	31.7
	2	16.9	13.8	15.5	32.8	24.9	24.1
	3	17.8	14.7	16.4	34.7	26.4	24.2
10 cm.	1	26.5	26.5	26.5	29.3	29.3	29.3
	2	21.3	18.3	19.0	25.8	21.4	20.3
	3	21.8	18.8	19.6	26.4	21.9	20.7
15 cm.	1	27.7	27.7	27.7	28.0	28.0	28.0
	2	23.6	20.9	20.5	23.7	20.6	20.1
	3	23.8	21.1	20.9	24.1	20.9	20.5
20 cm.	1	27.9	27.9	27.9	27.6	27.6	27.6
	2	24.3	21.9	21.1	23.8	21.0	20.7
	3	24.5	22.1	21.2	24.0	21.2	20.8

During the first week when all the plots were similar, the temperatures at all depths in the three plots were in agreement. The mean temperatures at the different depths during weeks No. 2 and No. 3 clearly bring out the effects of the watering alone in B and of watering and crop cover in C. In the morning, soil temperatures in plot C in the layers near the surface are warmer than in plots A or B. This is due to the blanketing effects of the vegetative cover which keeps even the air layers inside the vegetation warmer than those outside over bare ground. In the afternoon, however, plot C is cooler than plot A by as much as 26.5°C , 10.6°C , 8.7°C , 5.5°C , 3.6°C and 3.1°C at depths of 0, 2, 5, 10, 15 and 20 cm. The plot B is also cooler than plot A but is warmer than plot C in the afternoon. The conditions during week No. 3 are similar. The effect of a covering of vegetation in keeping down the temperatures during hours of insolation is well illustrated by these data.

EXPERIMENTS WITH SOIL BLOCKS

Experiment 1: Black cotton soil (control), Trivandrum sand and Sakrand soil blocks

In the present section we shall deal with experiments with blocks of some typical soils. These experiments were started with soils from Trivandrum (sand), and Sakrand (alluvium). Pits $6\frac{1}{2}$ ft. by $3\frac{1}{2}$ ft. and 1 ft. deep were dug, keeping the bottom of the pits horizontal. The vertical sides of the pits were supported with a lining of brick and cemented up. The lining of cement helps to prevent the seepage of water from the sides during rainy weather. These pits were carefully packed with the soils referred to above, the top surfaces of the different soil blocks so obtained being kept horizontal and at the same level as that of the ground in the neighbourhood. The sets of soil thermometers were then installed in these soil blocks and compared with those in the permanent control plot. The natural surfaces of the respective soil blocks were kept undisturbed during the first part of the experiment which extended from the 30th of April to 10th of May, 1936. The second part of the experiment was commenced on the 11th of May at 0800 hrs. when the blocks of Trivandrum sand and Sakrand soil were covered with a thin layer (2 mm.) of black cotton soil so as to equalize the surface colours and retain only the variation in the interior of the soils. The observations recorded during the first and the second part of the experiment have been averaged for the following periods both for 6 a.m. and 2 p.m.

No. of period	Dates	Remarks
1	30 April to 6 May 1936	} 1st part of experiment with each soil block having its own colour
2	7 May to 10 May 1936	
3	11 May to 20 May 1936	} 2nd part of experiment with all soils having a cover of black cotton soil
4	21 May to 27 May 1936	
5	28 May to 3 June 1936	
6	4 June to 10 June 1936	

Table X gives the mean soil temperatures at different depths for the above 6 periods at 6 a.m. and 2 p.m. From the table it will be seen that during the first two periods before equalizing the covers the temperatures in the upper layers of Trivandrum sand are lower than those in the control both in the morning and evening. The surface temperature in the Sakrand soil is lower than that in the control but higher than that in the Trivandrum sand both in the morning and in the afternoon ; but below 10 cm. the temperatures are slightly warmer than in the other two soils. After equalizing the covers, i.e. during the periods 3 to 6 the temperatures in the sand and in the Sakrand soil begin to increase rapidly and approach those in the control. Sand being a poor conductor of heat, the afternoon temperature just below the surface is higher in it than in either the control or the Sakrand soil ; for the same reason the morning temperatures in the uppermost layers of the sand are lower than in the other two cases in spite of the colours having been equalized.

TABLE X

Morning and afternoon average soil temperatures in °C in three soil blocks of Poona soil (control), Trivandrum sand and Sakrand soil during six weeks. Weeks 1 and 2 show the temperatures in the blocks with their natural surface colours ; during weeks 3, 4, 5 and 6 a thin cover of Poona soil equalized all the surface colours

No. of week	0600 hours			1400 hours		
	Control	Trivand- rum sand	Sakrand soil	Control	Trivand- rum sand	Sakrand soil
	A	D	E	A	D	E
0 cm.						
1 . . .	21.9	17.9	20.3	69.7	58.1	63.8
2 . . .	21.7	19.1	21.1	68.1	58.0	64.5
Thin 'covers' of Poona soil were applied to plots E and D at 0800 hrs. (11-5-1936).						
3 . . .	23.3	20.3	23.2	68.9	68.1	66.6
4 . . .	24.9	22.1	24.6	63.5	64.3	62.8
5 . . .	22.9	20.5	22.9	51.8	51.5	50.9
6 . . .	21.5	19.3	21.7	48.4	45.8	45.7
2 cm.						
1 . . .	26.7	24.1	24.4	54.9	53.8	55.8
2 . . .	28.5	24.7	25.3	52.1	52.5	54.9

TABLE X—*contd.*

No. of week	0600 hours			1400 hours		
	Control	Trivandrum sand	Sakrand soil	Control	Trivandrum sand	Sakrand soil
	A	D	E	A	D	E

Thin 'covers' of Poona soil were applied to plots D and E at 0800 hrs. (11-5-1936)

3 . . .	28.4	26.1	27.0	51.6	58.9	55.8
4 . . .	29.1	27.4	28.0	49.1	56.3	53.9
5 . . .	26.1	25.7	24.9	40.3	46.0	44.9
6 . . .	24.0	24.4	23.4	36.2	42.7	41.6

5 cm.

1 . . .	27.5	26.6	27.3	48.6	45.8	48.8
2 . . .	28.5	27.1	28.0	47.8	45.8	48.5

Thin 'covers' of Poona soil were applied to plots D and E at 0800 hrs. (11-5-1936)

3 . . .	28.9	28.8	29.5	46.3	50.0	49.2
4 . . .	29.5	29.3	30.0	44.1	49.0	47.8
5 . . .	26.1	27.3	26.9	37.0	40.5	39.7
6 . . .	24.1	26.1	25.3	32.6	38.2	37.3

10 cm.

1 . . .	31.5	29.2	31.3	38.1	39.9	40.0
2 . . .	32.2	29.9	31.9	38.1	39.7	40.3

Thin 'covers' of Poona soil were applied to plots D and E at 0800 hrs. (11-5-1936)

3 . . .	32.7	31.6	32.7	37.5	42.7	41.2
4 . . .	32.5	31.8	32.7	36.8	41.9	40.7
5 . . .	30.1	29.5	29.8	33.6	36.5	35.8
6 . . .	27.8	28.0	27.9	30.5	35.0	34.3

15 cm.

1 . . .	33.3	31.6		34.0	35.1	
2 . . .	33.8	32.2		34.4	35.3	

TABLE X—*concl'd.*

No. of week	0600 hours			1400 hours		
	Control	Trivand- rum sand	Sakrand soil	Control	Trivand- rum sand	Sakrand soil
	A	D	E	A	D	E

Thin 'covers' of Poona soil were applied to plots D and E at 0800 hrs. (11-5-1936)

3 . . .	34.0	34.0		34.4	37.7	
4 . . .	33.6	34.0		34.1	37.3	
5 . . .	31.9	31.5		32.0	34.0	
6 . . .	29.6	29.8		29.8	32.5	

20 cm.

1 . . .	33.3	33.2	34.5	33.0	33.1	33.9
2 . . .	33.7	33.7	34.9	33.3	33.4	34.4

Thin 'covers' of Poona soil were applied to plots D and E at 0800 hrs. (11-5-1936)

3 . . .	33.8	35.3	35.8	33.4	35.1	35.2
4 . . .	33.3	35.3	35.5	33.0	35.1	35.1
5 . . .	32.1	33.0	33.2	31.8	33.0	32.9
6 . . .	29.9	31.0	31.1	29.7	31.4	31.2

30 cm.

1 . . .	33.3	32.9	33.9	33.1	32.7	33.8
2 . . .	33.7	33.3	34.3	33.6	33.2	34.3

Thin 'covers' of Poona soil were applied to plots D and E at 0800 hrs. (11-5-1936)

3 . . .	33.8	34.5	35.1	33.6	34.2	34.9
4 . . .	33.3	34.5	35.0	33.2	34.3	34.7
5 . . .	32.3	33.6	33.6	32.1	33.3	33.3
6 . . .	30.1	32.0	31.9	30.1	31.7	31.6

Experiment 2 : Black cotton soil (control) and Bangalore (red) soil blocks

A similar experiment comparing the temperature in the control plot and in a block of red soil from Bangalore was started on the 9th of May 1936, as soon as a supply of the latter soil was received. The periods into which the 1st and 2nd parts of the experiment were divided were as follows :—

No. of period	Dates	Remarks
1	9 May to 15 May	} 1st part of experiment with each soil block having its own colour
2	16 to 22 May	
3	23 to 29 May	} 2nd part of experiment with Bangalore soil block having a cover of black cotton soil.
4	30 May to 5 June	
5	6 to 12 June	

Table XI gives the mean soil temperatures at different depths for the above 5 periods at 6 a.m. and 2 p.m. From the table it will be seen that during the first two periods before equalizing the covers the temperature at the surface of the Bangalore soil is lower than that of the control in the afternoon, but slightly greater than that of the control at lower depths. On applying the cover of the local soil at the beginning of the 3rd week, the afternoon surface temperature becomes more or less similar to that in the control but lower depths become still warmer owing to the larger absorption of energy at the surface and its conduction downwards. The changes of temperature on covering with local soil in the case of Bangalore soil are not of course so conspicuous as in the case of Trivandrum sand in the previous experiment. Further work on these lines is in progress at the observatory.

TABLE XI

Morning and afternoon average soil temperatures in °C in two soil blocks of Poona soil (control) and Bangalore soil during five weeks, weeks 1 and 2 show the temperatures in the blocks with their natural surface colours ; during weeks 3, 4 and 5 the surface colours are equalized in the blocks, a thin cover of Poona soil being applied to Bangalore soil block

Week No.	0600 hours		1400 hours	
	Control	Bangalore soil	Control	Bangalore soil
0 cm.				
1	21.7	22.1	69.5	65.3
2	24.9	24.7	66.6	62.6

TABLE XI—*contd.*

Week No.	0600 hours		1400 hours	
	Control	Bangalore soil	Control	Bangalore soil

Thin cover of Poona soil was applied at 1800 hours on 22nd May 1936.

3	24.8	25.1	59.7	60.1
4	22.2	22.6	45.8	45.4
5	21.3	21.9	53.5	52.0

5 cm.

1	28.6	27.4	47.4	50.1
2	29.3	28.5	45.4	48.1

Thin cover of Poona soil was applied at 1800 hours on 22nd May 1936.

3	29.3	28.8	42.5	47.5
4	24.7	24.9	33.7	36.4
5	24.7	25.4	34.4	40.5

10 cm.

1	32.6	31.0	38.1	43.0
2	32.6	31.1	37.1	41.8

Thin cover of Poona soil was applied at 1800 hours on 22nd May 1936.

3	32.2	31.6	36.4	41.5
4	28.9	27.5	31.7	35.0
5	28.1	27.7	31.2	37.8

15 cm.

1	34.1	34.0	34.6	36.6
2	33.9	33.6	34.3	36.1

Thin cover of Poona soil was applied at 1800 hours on 22nd May 1936.

3	33.5	34.0	33.7	35.9
4	30.9	30.3	31.0	32.4
5	29.8	30.2	30.1	33.4

The solution of the above equation for the case where the temperature at the surface of the soil undergoes a diurnal variation, which may be for simplicity represented by a sine curve, is given by

$$\theta = \theta_0 e^{\frac{-2\pi x}{\lambda}} \sin 2\pi \left(\frac{t}{T} - \frac{x}{\lambda} \right) \quad (2)$$

where T = time period of the wave, i.e., 24 hours and λ = 'wave-length', i.e. distance between points at which the maxima or minima of temperature occur simultaneously. By substituting the above solution in (1) we can

show that $k = \frac{\lambda^2}{4\pi T}$ so that

$$\frac{2\pi}{\lambda} = \sqrt{\frac{\pi}{Tk}} \quad (3)$$

If we put θ_0 = the amplitude of the diurnal variation at depth $x=0$, it is clear that the amplitude at a depth x is given by

$$\theta_0 e^{-\sqrt{\frac{\pi}{Tk}} \cdot x}$$

Knowing the amplitudes of diurnal variation at two depths x_1 and x_2 , we have from the above relation

$$\frac{\theta_1}{\theta_2} = e^{-(x_1 - x_2) \sqrt{\frac{\pi}{Tk}}} \quad (4)$$

Putting $T=24$ hours = 86,400 seconds, and taking logarithms of the various quantities to the base 10, we have

$$\log k = 2 \left\{ \log (x_1 - x_2) - \log (\log \theta_1 - \log \theta_2) \right\} - 5.1640 \quad (5)$$

Knowing x_1 , x_2 and θ_1 and θ_2 we can easily calculate k , the diffusivity of the soil at different intervals of depth.

Fig. 5 shows the diurnal variation of soil temperature on a clear day, viz. 0600 hrs. of the 30th April to 0600 hrs. of the 1st May 1935, at depths of 0, 2, 5, 10, 15 and 20 cms. below the surface. It will be noticed that besides the rapid decrease of the amplitude of the temperature wave with depth there is also a progressive lag in the epochs of maximum and minimum temperature. This is easily understood from equation (2). The maximum

temperature at a depth x_1 is attained at time t_1 , when $\sin 2\pi \left(\frac{t_1}{T} - \frac{x_1}{\lambda} \right) = 1$

$$\text{or where } 2\pi \left(\frac{t_1}{T} - \frac{x_1}{\lambda} \right) = \left(2n + \frac{1}{2} \right) \pi$$

Similarly the maximum epoch at a depth x_2 will be attained at time t_2 ,

$$\text{given by } 2\pi \left(\frac{t_2}{T} - \frac{x_2}{\lambda} \right) = \left(2n + \frac{1}{2} \right)$$

By subtraction we have

$$2\pi \left(\frac{t_2 - t_1}{T} - \frac{x_2 - x_1}{\lambda} \right) = 0$$

so that

$$\frac{t_2 - t_1}{x_2 - x_1} = \frac{T}{\lambda} = \sqrt{\frac{T}{4\pi T k}} = \sqrt{\frac{T}{4\pi k}}$$

We can thus find out k from the variation of the phase of the maximum or minimum temperature epochs, and put

$$k = \frac{T}{4\pi} \cdot \frac{(x_2 - x_1)^2}{(t_2 - t_1)^2} \dots\dots\dots (6)$$

or $\log k = 3.8373 + 2 \log (x_2 - x_1) - 2 \log (t_2 - t_1) \dots\dots\dots (7)$,
the logarithms being taken to the base 10.

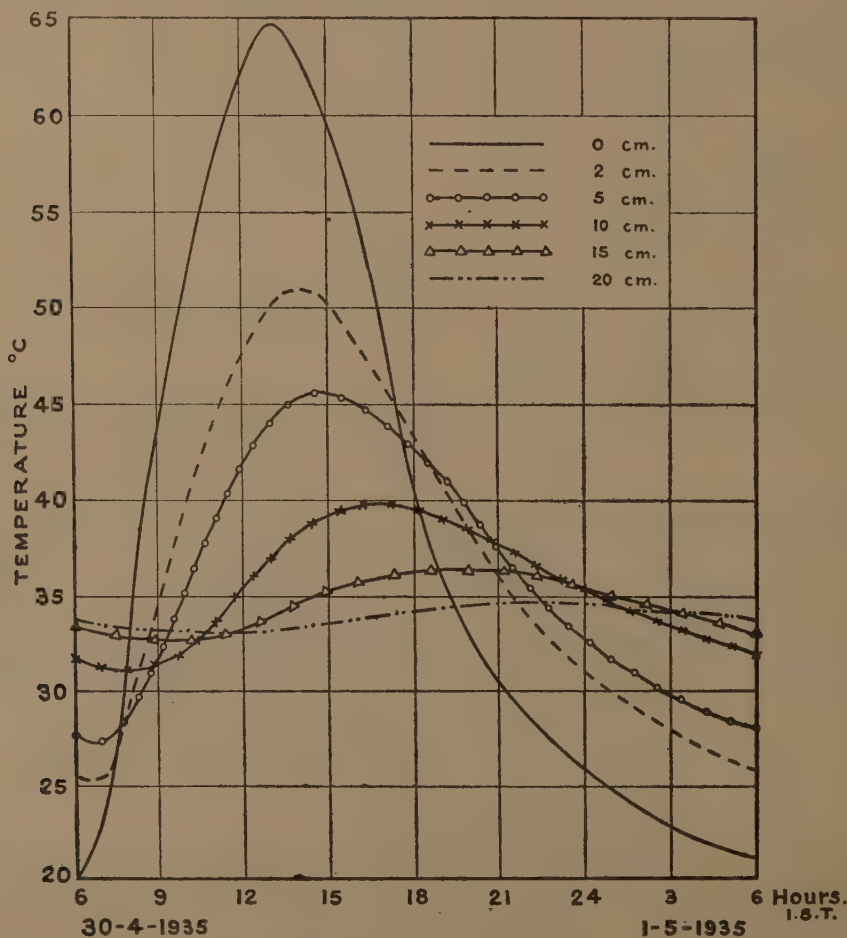


FIG. 5. Diurnal variation of soil temperature at 0, 2, 5, 10, 15 and 20 cm. depths from 0600 hrs. of 30-4-1935 to 0600 hrs. of 1-5-1935

For calculating k from the temperature curves, it is more convenient to use equation (5) as the amplitudes of the curves are much easier to determine than the epochs of maximum and minimum temperatures. This difficulty increases with depth, as the curves become flatter and flatter.

Variation of thermal diffusivity with depth in Poona soil

Table XII gives the amplitudes of soil temperature and the times at which the maximum and minimum temperatures occur at different depths. The last two columns give the values of the thermal diffusivity k calculated from the amplitudes and the values of K the thermal conductivity obtained by multiplying k by the product of the apparent density and the specific heat. The apparent density varies with depth as follows, but the specific heat has a constant value of 0.22.

Depth (cm.)	Apparent density			
0	.	.	.	1.0
5	.	.	.	1.9
10	.	.	.	2.1
15	.	.	.	2.3
20	.	.	.	2.3

The thermal conductivity is of the order of 0.0004 at the surface but increases to values lying between 0.0005 and 0.0007, at lower depths, owing to the increase of the apparent density at these depths.

TABLE XII

Depth	Amplitude	Maximum temperature epoch	Minimum temperature epoch	Diffusivity k (calculated from amplitude)	Conductivity K
0 cm.	22.4	1300	0600	0.0011	0.00037
5 cm.	9.2	1500	0700	0.0015	0.00067
10 cm.	4.3	1700	0800	0.0012	0.00060
15 cm.	1.8	2000	0930	0.0010	0.00052
20 cm.	0.7	2230	1130		

Seasonal variation of thermal diffusivity

We may now see how the thermal diffusivity of the soil at Poona varies with the season. The mean soil temperatures at different depths from 0 to 50 cm. for different months during the period April 1936 to March 1937 were computed for 0600 and 1400 hours which are the epochs of minimum and maximum soil temperatures respectively at the surface and for 0800 and 1700 hours which are the epochs of minimum and maximum soil temperatures

respectively at a depth of 10 cm. From these the diurnal ranges of soil temperatures for the depths of 0 and 10 cm. were found and the mean thermal diffusivity of the surface layer of the soil, 10 cm. in thickness, calculated. From the records of the Central Agricultural Meteorological Observatory, we know also the rainfall and mean moisture content of this surface layer. The values of diffusivity and of the rainfall and mean moisture content of the first 10 cm. of the soil are given in Table XIII.

TABLE XIII

Monthly variation of diffusivity (between the depths of 0 and 10 cm).

Month	Rainfall in inches	k	Moisture content (per cent on dry basis)
April 1936	0.00	0.0016	7.5
May 1936	0.56	0.0015	7.5
June 1936	3.82	0.0013	12.0
July 1936	1.47	0.0023	25.0
August 1936	1.34	0.0018	13.0
September 1936	7.28	0.0024	25.0
October 1936	0.88	0.0018	15.0
November 1936	2.99	0.0023	21.0
December 1936	0.00	0.0013	13.4
January 1937	0.00	0.0013	8.7
February 1937	0.00	0.0015	7.7
March 1937	0.00	0.0016	5.3

Mean k during the dry season December to May : 0.0015

Mean k during the wet season June to November : 0.0020

During the months December to May there is practically no rainfall and the mean moisture content of the soil between the surface and 10 cm. depth lies between 5 and 13 per cent ; consequently the values of thermal diffusivity also are low, ranging between 0.0013 to 0.0016. The mean diffusivity of the first 10 cm. of the soil during these six dry months of the year is 0.0015. The monsoon sets in in June and the remaining six months of the year are wet. The mean moisture content of the soil is seen to increase up to 25 per cent and the diffusivity from 0.0013 to 0.0024. The mean value of the diffusivity during these six wet months is 0.002.

June has a rainfall of 3·82 in. and yet the value of the diffusivity is seen to be low (0·0013). But we note that the mean moisture content of the soil is also low during this month, viz. 12 per cent, the rainfall having occurred at the end of June. The heavy rainfall at the end of June and further rainfall in July raise the value of the mean moisture content during July to 25 per cent and the diffusivity is also seen to attain the high value of 0·0023 during this month. In September and November again heavy rainfall is recorded, the moisture content of the soil rises in value, and the diffusivity is also seen to be high.

We find therefore that in general the thermal diffusivity of the soil varies directly with its moisture content. This must be attributed to the fact that with increase of moisture, water fills the inter-space between the soil particles, driving away the air which has a low thermal diffusivity.

Thermal diffusivity in relation to surface colour

Table XIV gives the values of thermal diffusivity for the plots A, B, C, D and E during the week January, 8th to 14th, 1935, when all these plots of local Poona soil were exposed to solar radiation with their natural surface colours. The plots A, B, C, D and E have the values of 0·0013, 0·0013, 0·0014, 0·0012 and 0·0013 respectively. The table also gives the values of diffusivity for the plots as 0·0013, 0·0014, 0·0014, 0·0012 and 0·0012 respectively during the week January, 29th to February, 4th, 1935, when the plots B, C, D and E were exposed to solar heating with their surface colours changed by the covers of Trivandrum sand (white), Mekran soil (brown), Sakrand soil (ash-coloured) and Bangalore soil (red), the covers having been applied at 0700 hours on 24th of January 1935. Here we find that all the five plots consisting of the local Poona soil show about the same value of diffusivity whatever be the surface colour, showing that other conditions remaining the same mere change of albedo at the surface makes no alteration in the thermal conductivity in the soil below.

TABLE XIV

Week	Plot	<i>k</i>
1935—January 8—14	A	0·0013
	B	0·0013
	C	0·0014
	D	0·0012
	E	0·0013
January 24th to February, 4th	A	0·0013
	B	0·0014
	C	0·0014
	D	0·0012
	E	0·0012

Variation of thermal diffusivity with soil types

Table XV gives the thermal diffusivity values for the local Poona soil, Trivandrum sand and Sakrand soil, both when the blocks of these soils were exposed to solar heating with their natural surface colours and when their surface colours were equalized by means of a thin cover of the Poona soil.

TABLE XV

Diffusivity—(between the depths of 0 and 10 cm.)

Soil	Before surface treatment	After the surface colours were equalized
8 May 1936		
Poona soil	0·0014	0·0011
Trivandrum sand	0·0036	0·0032
Sakrand soil	0·0027	0·0024
20 May 1936		
Poona soil	0·0011	0·0011
Bangalore soil	0·0035	0·0031

Poona soil, Trivandrum sand and Sakrand soil have the diffusivity values of 0·0014, 0·0036 and 0·0027 respectively in their natural condition; and they do not show much difference after the surface treatment.

The table also gives the diffusivity values of 0·0011 and 0·0035 for the Poona soil and Bangalore soil respectively before the soil blocks received the surface treatment. These values are also not affected by the change in the surface colour.

Summary and conclusion

The present paper begins with a description of Poona and its environs and climate (Section I). In the next section the various factors which control the thermal balance at the surface of the ground during the clear season are briefly mentioned. Some typical measurements of the intensity of the radiation from the sun and the sky, the albedo factor which determines the fraction of the radiation actually absorbed by the soil surface, the heat transfer at the surface by conduction, convection and by radiative exchange in the infra-red region of the spectrum are briefly discussed in this section (Section II).

Section III is devoted to an outline of the scheme of experiments discussed later in the paper. In Section IV five experiments on the influence of surface 'covers' on soil temperatures are described. These experiments show how sensitive soil temperatures are to changes of colour or surface

wetness. In many investigations of plant physiologists and agriculturists it is often necessary to alter soil temperatures to suit the requirements of a crop. For example, in higher latitudes where the intensity of solar radiation is low it becomes necessary to make the best use of the weak insolation. In Soviet Russia, for example, the method of covering the soil surface with charcoal powder or coal dust is reported to have been tried with success for making the soil temperatures sufficiently high to sustain a cotton crop. In the hot Indian summer we may often have to keep down soil temperatures to save plants during a droughty period. The use of a thin layer of a white substance like chalk is obvious in such circumstances. The control of soil temperatures by altering the soil cover will be possible only for depths up to 50 cm. or so, as the changes occurring at the surface decrease rapidly with depth.

Section V is devoted to experiments with blocks of different soils which show how the influence of the surface colour may be eliminated by covering all the soils with a thin layer of Poona soil.

The diurnal variations of soil temperatures, the thermal diffusivity of the various soils used in the previous experiments, and the seasonal variation of the thermal diffusivity of Poona soil are discussed in Section VI. It is shown that the diffusivity is not influenced by the surface treatments but increases when the moisture content of the interior of the soil is increased during the wet season.

The investigations discussed in the present paper were conducted under the guidance of Dr L. A. Ramdas, Agricultural Meteorologist, Meteorological Office, Poona. The present writer is grateful to Dr L. A. Ramdas for the suggestion of the problem and to Dr C. W. B. Normand, Director-General of Observatories, for the facilities given for the work at the Meteorological Office, Poona.

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A STUDY ON PLOT SIZE AND SHAPE TECHNIQUE FOR FIELD EXPERIMENTS ON SUGARCANE

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INTRODUCTION

SINCE the inception of the Sugarcane Research Scheme at Padegaon, financed by Imperial Council of Agricultural Research, experiments have been laid down on modern lines of field technique propounded by Dr. Fisher. To suit the local conditions, an experimental gross plot size of 54.45 ft. \times 32 ft. = 4 cents (4/100 acre) was considered convenient for experiments in this farm. This plot size covered eight rows of cane 4 ft. apart and omitting one row on either side of the plot for border effect, and 4.5 ft. on either end a net plot of 2.5 cents (i.e. 1/40th of an acre) was obtained. The plant population varied from 600 to 1000 in this unit plot-size depending on the variety. The main conditions of soil and growing of the crop in this area is as follows, which differ from conditions of dry crop in Northern India :—

(1) *Topography*.—The soil is extremely undulating and is found to have a gradient varying from 1 in 100 to 1 in 300 on both sides which is not found in the United Provinces or Bihar where gradients are 1 in 1,000 to 2,000.

(2) *Cane growing*.—Cane is required to be irrigated throughout the year at intervals of ten days and hence cane is planted 4 ft. apart. Regular and even distribution of irrigation water is important for this crop.

After one year's experience with the above plot size, the distribution of water with varying gradients in the farm was found to be regular and as a result crop growth was also found perfectly uniform. The uniform growth of the crop in a plot resulting from even distribution of irrigation water removed one cause of variability and gave satisfactory results.

Still it was thought necessary to conduct a special uniformity trial to have convincing proof about the soundness of the plot size already laid down and to see whether the plot size could not be further reduced, which information would be useful in laying out future experiments.

MATERIAL

Two areas 'A' and 'B' were planted with Co 360 cane in the first week of March 1934. A short description of the two plots showing the details of topography cultural operations, etc. is given below :—

	Block A	Block B	Remarks
(1) Size . . .	E-W 192 ft. × S-N 253·4 ft. equivalent to 1 acre 4·6 gunthas	E-W 132 ft. × N-S 483·4 ft. equivalent to 1 acre 18·6 gunthas.	Variety Co 360 (flowering)
(2) Topography .	Depth of soil 9 in. to 12 in. Depth of sub-soil 8 in. to 9 in. Lower strata of porous murum well drained	Depth of soil 9 in. to 12 in. Depth of sub-soil 8 in. to 9 in. Lower strata of porous murum well drained	
(3) Method under which cane was grown :—			
(1) Time of planting	7 March 1934 . . .	5 March 1934	
(2) Distance between rows	4 feet	4 feet	
(3) Seed rate .	10,000 sets of three eye-buds each per acre.	10,000 sets of three eye-buds each per acre	
(4) Number of irrigations	33	35	
(5) Manuring .	Farmyard manure at 25 carts per acre applied on 19th February 1934. 150 lb. of nitrogen was applied as top dressing in the form sulphate of ammonia and cake in the usual Manjri standard method.	Farmyard manure at 25 carts per acre applied on 17th February 1934. 150 lb. of nitrogen was applied as top-dressing in the form of sulphate of ammonia and cake in the usual Manjri standard method.	
(6) Earthing up	The crop was hand weeded and earthing up was done by a plough on 1st July 1934.	The crop was hand weeded and earthing up was done by a plough on 2nd July 1934	
(7) Date of harvest.	The crop was cut in strips of 10 ft. from 21st January 1935 to 26th January 1935.	The crop was cut in strips of 10 ft. from 4th February 1935 to 12th February 1935.	
(8) Yield in tons	35·97 per acre . . .	26·6 per acre	

For purposes of this study after discarding guard rows all round, 32 rows each 240 feet long were taken from plot A, and 30 rows each 400 feet long from plot B. The rows were cut in sections of 10 feet length. Thus there were 768 ultimate units (10 ft. \times 4 ft.) from plot A and 1200 units (10 ft. \times 4 ft.) from plot B.

STATISTICAL ANALYSIS

The distribution of yields from unit plots from both the blocks were found to be nearly normal as the following constants show :—

$$\text{PLOT A : } g_1 = -0.1059 \pm 0.0839$$

$$g_2 = 0.861 \pm 0.167$$

$$\text{PLOT B : } g_1 = 0.064 \pm 0.068$$

$$g_2 = -0.050 \pm 0.137$$

The 768 units of plot 'A' were grouped to give plots one row, two rows, four rows and eight rows wide and 10 ft., 20 ft., 30 ft., 40 ft., 60 ft., 80 ft. and 120 ft. long. The plots so formed were then grouped together to form four plot and eight plot (see below) blocks in different ways.

In the case of 1,200 units of plot B, the unit plots were grouped to give plots one row, two rows, three rows, five rows, six rows and ten rows wide and 10 ft., 20 ft., 40 ft., 50 ft., 80 ft. and 100 ft. long. Blocks were now formed by taking five plots together across rows and along rows.

In all cases the usual method of analysis of variance was adopted to remove the variation between blocks and thence to calculate the percentage standard error per plot for the particular case considered. In almost all cases there was an appreciable reduction in the variance by this process of elimination of major soil differences.

Table I gives the percentage standard error per plot (coefficient of variation) for both the fields for the different kinds of blocks. For field A, four types of blocks have been considered, viz. four-plot blocks across rows (i.e. the plots lying in a line at right angles to the direction of the rows), four plot blocks (in a line) along the direction of rows, four-plot blocks in two rows (compact), and eight plot blocks (compact only). In the last case the blocks have been taken in as compact a form as possible.

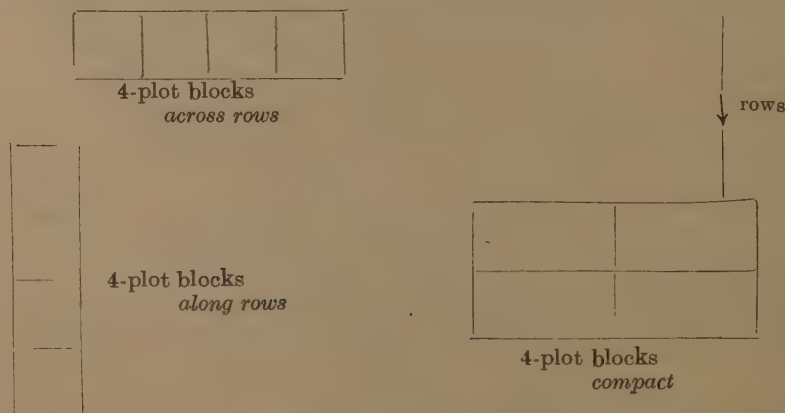


Diagram showing types of blocks.

For field B, five-plot blocks have been taken along rows and across rows

As has been found usual in similar analyses with other crops as well as sugarcane (for a fairly complete bibliography on uniformity trial data see Cochran [1937] and in addition Hutchinson and Panse [1935] and Macdonald *et al* [1939]), there is a gradual decrease in the values of the percentage error per plot by an increase either in the length of the rows or by the inclusion of greater number of rows (i.e. by increasing the width of the plot). It is seen that the reduction in variation is more rapid by increasing the length of rows. There is also a definite advantage in the inclusion of greater number of rows per plot up to say between four and eight rows. In the latter case it is however evident that there is no proportionate gain in precision; for instance in doubling the size of plot from four rows to eight rows, the precision is not increased two-fold. These tables indicate that the usual plot size adopted at the station (vide introduction) is quite satisfactory from the point of view of precision, and any reduction in the size would diminish the accuracy of the comparisons.

As regards the method of formation of blocks for removing major differences in soil fertility, these tables show that in the case of small plots there is very little to choose as to whether the blocks should be taken across rows, along rows or compact. In the case of bigger plots (of the type usually adopted in field experiments) there seems to be a slight advantage in taking the plots across the direction of rows. In field B however, five plot blocks along the direction of rows appeared better than across the rows.

There is also very little difference in the standard of accuracy by having eight plot blocks or four plot blocks as far as the present data are concerned.

In Table II plots of the same shape (i.e. length : breadth ratio) are grouped together and in Table III plots of the same size, but of different shapes. For any particular shape, the bigger the plot, the less the variation within the limits studied in this paper.

Table III shows that contrary to the usual, amongst plots of a given size up to about 1/90 acre, the longer plots appear to vary more than the shorter plots, and also when the plots are widened by increasing the number of rows (the width being greater than the length) the variation is as high as in the case of long plots. When the plot size is greater than 1/90 acre, the longer plots, in general, show less variation than the shorter plots. As the usual experimental plots fall within this range, the conclusion that fairly long plots are preferable to square plots seems to be supported by the present data. There seems to be also no advantage in having a large number of short rows per plot for sugarcane field experiments, i.e. for a given plot shape it is seen that it is more advantageous to have the longer dimension of the plot along the direction of rows than across. Similar results were obtained by Hutchinson and Panse [1935] working with Malvi mass-selected cotton data at Indore.

Table IV gives the number of replications and area of land required to give a standard error of 2 per cent of the mean. This means that differences of over 6 per cent of the mean can be taken to be significant ($P = 0.05$). The number of replications and area of land required is of course dependent on the standard of accuracy required. When a smaller degree of accuracy is sufficient (say differences of 12 per cent of the mean), standard error has

only to be 4 per cent of the mean and the number of replications will be half of what it is in Table IV.

A comparison of Tables IV A and IV B shows that the standard error and consequently the number of replications and area of land required for a given degree of accuracy are largely dependent on the amount of heterogeneity present. Field B as a whole shows greater variation for the whole range of plot sizes considered and hence the number of replications is also found to be comparatively high.

EFFICIENCY PER UNIT AREA OF LAND

Table V gives the relative efficiencies of plots in the use of land. The usual method has been employed in the calculation of these figures. The efficiency of a particular plot has been obtained by multiplying the percentage variance per plot by the number of units used to make up the plot ('unit' referring to the smallest plot size used in the present data), and taking the reciprocal. The smaller plots are seen to be comparatively more efficient than bigger plots although from the point of view of agricultural convenience no one would lay out experiments with too small plots. From field A, (four plot blocks) it will be seen that two-row plots 60 ft. to 80 ft. long are almost as efficient as the smallest-sized plots. When blocks are taken across rows, plots of 80 ft. length and eight rows width also seem to be quite efficient. With eight plot blocks, however there seems to be no advantage in increasing the number of rows per plot beyond four. Here also the optimum length of rows appears to be 60 to 80 ft. which is somewhat longer than the usual plot length. On the basis of these figures also it appears that the usual plot size adopted in this station is fairly satisfactory.

Field B shows that with five plot blocks the efficiencies of the bigger-sized plots are comparatively very much lower. As a matter of fact, in this area five plot blocks and ten plot blocks on the whole gave much higher standard errors as compared to blocks with two, four etc. plots. To study the influence of increasing the size of block, keeping the same plot size, standard errors were calculated separately for plot of size 40 ft. \times 20 ft. and the results are given below :—

Number of plots per block	Per cent standard error
2 . .	11.86
3 . .	11.10
4 . .	11.66
5 . .	17.08
6 . .	12.25
10 . .	16.48
12 . .	12.72

This shows that for some particular numbers of plots per block, the precision may be high, the optimum number depending on the nature of the field itself. In this field, there is very little difference between two, three or four plot blocks or even six or twelve plot blocks. Only five or ten plot blocks appear to show higher variation. This is an interesting conclusion, especially as the

number of plots per block has assumed a new importance with the advent of the 'incomplete block systems of lay-out' the quasi-factorial as well as the symmetrical types.

SUMMARY

The results of a uniformity trial on sugarcane conducted in two fields at Padegaon have been studied mainly with a view to examine the satisfactoriness of the usual plot size and shape adopted at the station, viz. (54.44 ft. \times 32 ft.) (eight rows 4 ft. apart) and it is found that under the conditions prevailing here, there seems to be no need for any appreciable change.

2. When the plot size is greater than 1/90 acre, in general, the longer plots show less variation than shorter plots. That fairly long plots are preferable to square plots is thus shown by the present data.

3. When four plot blocks are considered, it is found that two-row plots 60 ft. to 80 ft. long are almost as efficient as the smallest sized plots. When blocks are taken across rows, plots of 80 ft. length and eight rows width also seem quite efficient.

4. With eight plot blocks there appears to be no advantage in increasing the number of rows per plot beyond four.

5. It is also seen that there need be no restriction in the number of plots per block which might vary from two up to twelve depending upon the experimental treatments.

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TABLE I

Standard errors per cent for plots of different sizes and hap's

Row lengths in feet	Number of rows			
	1	2	4	8

Field A.—4-plot blocks across rows

10	26.00	18.13	15.13	12.44
20	19.53	12.94	9.59	7.55
30	17.04	12.05	9.00	6.29
40	15.92	11.18	7.18	6.26
60	12.11	7.90	6.33	5.24
80	9.76	6.66	5.69	3.40
120	9.15	6.27	4.85	..

Field A.—4-plot blocks along rows

10	25.64	18.59	15.34	12.28
20	19.65	13.37	9.90	7.46
30	17.09	12.14	9.25	6.43
60	12.05	9.01	7.21	5.57

Field A.—4-plot blocks compact

10	27.22	18.54	15.25	12.67
20	19.58	13.05	9.24	7.38
30	17.52	12.11	8.91	6.43
40	16.19	12.11	7.27	6.01
60	12.15	8.27	6.33	5.43
120	9.83	7.15	5.01	..

TABLE I—*contd.*

Row lengths in feet	Number of rows			
	1	2	4	8
Field A.—8-plot blocks compact				
10	27.02	18.52	14.96	10.40
20	19.38	12.96	9.75	8.05
30	17.22	12.05	9.12	6.76
40	15.48	9.67	7.88	6.74
60	12.04	8.27	6.68	5.59
80	9.61	7.42	5.74	..
120	9.04	6.63	5.11	..

Row lengths in feet	Number of rows					
	1	2	3	5	6	10
Field B.—5-plot blocks across rows						
10	31.44	23.55	24.10	16.83	15.17	12.57
20	27.98	18.82	16.51	13.65	12.22	10.25
40	24.24	19.30	18.28	17.08	15.76	15.40
50	*15.79	..	*15.04
80	21.41	16.89	15.88	14.97	14.30	13.81
100	*15.05	..	*14.32

Field B.—5-plot blocks along rows					
10	36.20	24.13	24.12	..	12.30
20	29.19	18.95	16.37	..	11.81
40	21.38	16.20	12.88	..	9.34
50	20.20	13.62	11.55	†8.50	8.88
80	18.05	12.03	10.57	..	8.38
100	16.01	10.88	8.65	†14.81	7.07

* 4-plot Blocks (across).

†4-plot Blocks (compact).

TABLE II-A

(Field A)

Standard errors per cent of plots of different sizes arranged according to shape

Plot shape (length : breadth)	Plot size (acre)	4-plot block			8-plot blocks
		across rows	along rows	compact	
2.5 : 1	1/1089	26.00	25.64	27.22	27.02
	1/272	12.94	13.37	13.05	12.96
	1/68	7.18	..	7.27	7.88
	1/17	3.40
5 : 1	1/544	19.53	19.65	19.58	19.38
	1/36	11.18	..	12.11	9.67
	1/34	5.69	5.74
7.5 : 1	1/363	17.04	17.09	17.52	17.22
	1/90	7.90	9.01	8.27	8.27
	1/22	4.85	..	5.01	5.11
10 : 1	1/272	15.92	..	16.19	15.48
	1/68	6.66	7.42
15 : 1	1/181	12.11	12.05	12.15	12.04
	1/45	6.27	..	7.15	6.63
20 : 1	1/36	9.76	9.61
30 : 1	1/91	9.15	..	9.83	9.04
1.25 : 1	1/544	18.13	18.59	18.54	18.52
	1/136	9.59	9.90	9.24	9.75
	1/34	6.26	..	6.01	6.74
3.75 : 1	1/181	12.05	12.14	12.11	12.05
	1/45	6.33	7.21	6.33	6.68
	1/11
6.25 : 1	1/272	15.13	15.34	15.25	14.96
	1/68	7.55	7.46	7.38	8.50
1.875 : 1	1/90	9.00	9.25	8.91	9.12
	1/22	5.24	5.57	5.43	5.59
3125 : 1	1/136	12.44	12.28	12.67	10.40
9375 : 1	1/45	6.29	6.43	6.43	6.76

TABLE II-B

(*Field B*)

Standard errors per cent of plots of different sizes arranged according to shape

Plot shape (length : breadth)	Plot size (acre)	5-plot blocks	
		across	along
25 : 1	1/109	..	16·01
20 : 1	1/136	21·41	18·05
12·5 : 1	1/218	..	20·20
10 : 1	1/272	24·24	21·38
	1/68	16·89	12·03
5 : 1	1/544	27·29	29·19
	1/136	19·30	16·20
	1/22	*15·05	†14·81
2·5 : 1	1/1089	34·44	36·20
	1/272	18·82	18·95
	1/44	*15·79	†8·50
	1/11	*14·31	..
1·25 : 1	1/544	23·55	24·13
	1/22	*15·04	..
6·25 : 1	1/109	..	13·62
12·5 : 1	1/54	..	10·88
·5 : 1	1/218	16·83	..
	1/105	10·25	..
·25 : 1	1/109	12·57	..
·83 : 1	1/363	24·10	24·12
	1/90	12·22	11·81
1·7 : 1	1/181	16·51	16·37
	1/45	15·76	9·34
3·3	1/90	18·28	12·88
	1/22	14·30	8·38
4·2 : 1	1/73	..	11·55
	1/18	..	7·07
6·7 : 1	1/45	15·88	10·57
8·3 : 1	1/36	..	8·65
1 : 1	1/109	13·65	..
	1/27	15·40	..
2 : 1	1/54	17·08	..
	1/13	13·81	..
4 : 1	1/27	14·97	..
2 : 1	1/36	..	8·88
·4 : 1	1/181	15·17	12·30

*4-plot blocks (across).

† 4-plot blocks (compact).

TABLE III-A

(Field A)

Standard errors per cent of plots of different shapes arranged according to size

Plot size	Plot shape (length : breadth)	4-plot blocks			8-plot blocks
		across rows	along rows	compact	
1/1089	2.5 : 1	26.00	25.64	27.22	27.02
1/544	5 : 1	19.53	19.65	19.58	19.38
	1.25 : 1	18.13	18.59	18.54	18.52
1/363	7.5 : 1	17.04	17.09	17.52	17.22
1/272	10 : 1	15.92	..	16.19	15.48
	2.5 : 1	12.94	13.37	13.04	12.96
	.625 : 1	15.13	15.34	15.25	14.96
1/181	15 : 1	12.11	12.05	12.15	12.04
	3.75 : 1	12.05	12.14	12.11	12.05
1/136	20 : 1	9.76	9.61
	5 : 1	11.18	..	12.11	9.67
	1.25 : 1	9.59	9.90	9.24	9.75
	.3125 : 1	12.44	12.28	12.67	10.40
1/90	30 : 1	9.15	..	9.83	9.04
	7.5 : 1	7.90	9.01	8.27	8.27
	1.875 : 1	9.00	9.25	8.91	9.12
1/68	10 : 1	6.66	7.42
	2.5 : 1	7.18	..	7.27	7.88
	.625 : 1	7.55	7.46	7.38	8.50
1/45	15 : 1	6.27	..	7.15	6.63
	3.75 : 1	6.33	7.21	6.33	6.68
	.9375 : 1	6.29	6.43	6.43	6.76
1/34	5 : 1	5.69	5.74
	1.25 : 1	6.26	..	6.01	16.74
1/22	7.5 : 1	4.85	..	5.01	5.11
	1.875 : 1	5.24	5.57	5.43	5.59
1/17	2.5 : 1	3.40
1/11	3.75 : 1

TABLE III-B

(Field B)

Standard errors per cent of plots of different shapes arranged according to size

Plot size (acre)	Plot shape (length : breadth)	5-plot blocks	
		across rows	along rows
1/1089	2.5 : 1	34.44	36.20
1/544	5 : 1	27.98	29.19
	1.25 : 1	23.55	24.13
1/363	.83 : 1	24.10	24.12
1/272	10 : 1	24.24	21.38
	2.5 : 1	18.82	18.95
1/218	12.5 : 1	..	20.20
	0.5 : 1	16.83	..
1/181	1.7 : 1	16.51	16.37
	.4 : 1	15.17	12.30
1/136	20 : 1	21.41	18.05
	5 : 1	19.30	16.20
1/109	25 : 1	..	16.01
	6.25 : 1	..	13.62
	1 : 1	13.65	..
	.25 : 1	12.57	..
1/105	0.5 : 1	10.25	..
1/90	3.3 : 1	18.28	12.88
	0.83 : 1	12.22	11.81
1/73	4.2 : 1	..	11.55
1/68	10 : 1	16.89	12.03
1/54	12.5 : 1	..	10.88
	2 : 1	17.08	..
1/45	6.7 : 1	15.88	10.57
	1.7 : 1	15.76	9.34
	2.5 : 1	*15.79	†8.50
1/36	8.3 : 1	..	8.65
	2.1 : 1	..	8.88
1/27	4 : 1	14.97	..
	1 : 1	15.40	..
1/22	5 : 1	*15.05	†14.81
	3.3 : 1	14.30	8.38
	1.25 : 1	*15.04	..
1/18	4.2 : 1	..	7.07
1/13	2 : 1	13.81	..
1/11	2.5 : 1	*14.31	..

* 4-plot blocks (across). † 4-plot blocks (compact).

(Field A)

Number of replications and area of land required to give a standard error of 2 per cent of the mean

[illegible]

TABLE IV-B

(Field B)

Number of replications and area of land required to give a standard error of 2 per cent of the mean

Plot size (acre)	Plot shape (length : breadth)	Number of replications (5-plot blocks)		Total area	
		Across rows	Along rows	Across rows	Along rows
1/1089	2.5 : 1	296	328	1.36	1.50
1/544	5 : 1	196	213	1.80	1.96
	1.25 : 1	139	145	1.28	1.33
1/363	0.83 : 1	145	145	2.00	2.00
1/272	10 : 1	147	114	2.70	2.09
	2.5 : 1	88	90	1.62	1.65
1/218	12.5 : 1	..	102	..	2.34
	0.5 : 1	71	..	1.63	..
1/181	1.7 : 1	68	67	1.88	1.85
	0.4 : 1	57	38	1.57	1.05
1/136	20 : 1	114	81	4.19	2.98
	5 : 1	93	66	3.42	2.43
1/109	25 : 1	..	64	..	2.93
	6.25 : 1	..	46	..	2.11
	1 : 1	46	..	2.11	..
	0.25 : 1	39	..	1.79	..
1/105	0.5 : 1	26	..	1.24	..
1/90	3.3 : 1	83	41	4.61	2.28
	0.83 : 1	37	35	2.05	1.94
1/73	4.2 : 1	..	33	..	2.26
1/68	10 : 1	71	36	5.22	2.65
1/54	12.5 : 1	..	29	..	2.68
	2 : 1	73	..	6.76	..
1/45	6.7 : 1	63	28	7.00	3.11
	1.7 : 1	62	22	6.89	2.44
	2.5 : 1	*62	†18	5.51	1.60
1/36	8.3 : 1	..	19	..	2.64
	2.1 : 1	..	20	..	2.78
1/27	4 : 1	56	..	10.37	..
	1 : 1	59	..	10.92	..
1/22	5 : 1	*57	†55	*10.36	†10.00
	3.3 : 1	51	17	11.59	4.86
	1.25 : 1	*56	..	*10.18	..
1/18	4.2 : 1	..	12	..	3.33
1/13	2 : 1	48	..	18.46	..
1/11	2.5 : 1	*51	..	*18.54	..

*4-plot blocks (across).

†4-plot blocks (compact.)

TABLE V
Efficiency of plots in use of land

Row lengths in feet	Number of rows			
	1	2	4	8
<i>Field A.—4-plot blocks across rows</i>				
10 . .	100	102	98	55
20 . .	88	101	92	74
30 . .	77	77	69	71
40 . .	66	67	82	54
60 . .	76	90	70	51
80 . .	89	95	65	91
120 . .	67	72	60	..
<i>Field A.—4-plot blocks along rows</i>				
10 . .	100	95	69	54
20 . .	85	92	84	74
30 . .	75	74	64	66
40
60 . .	75	67	53	44
80
120
<i>Field A.—4-plot blocks compact</i>				
10 . .	100	107	79	58
20 . .	96	108	108	85
30 . .	80	84	78	75
40 . .	70	63	87	64
60 . .	83	90	77	52
80
120 . .	64	60	63	..

TABLE V—*contd.*

Row lengths in feet	Number of rows			
	1	2	4	8
<i>Field A.—8-plot blocks compact</i>				
10	100	106	81	84
20	97	109	96	70
30	82	84	73	66
40	76	97	73	50
60	84	89	68	49
80	99	83	69	..
120	74	69	58	..

Row lengths in feet	Number of rows					
	1	2	3	5	6	10
<i>Field B.—5-plot blocks across rows</i>						
10	100	107	68	84	86	75
20	76	84	72	64	66	56
40	50	40	29	20	20	12
50	*19	..	*10
80	32	26	19	13	12	8
100	*10	..	*6

<i>Field B.—5-plot blocks along rows</i>						
10	100	112	75	..	144	
20	77	91	81	..	78	
40	72	62	66	..	62	
50	64	71	65	†72	55	
80	50	56	49	..	39	
100	51	55	58	†12	44	

*4-plot blocks (across).

†4-plot blocks (compact).

INTERSPECIFIC HYBRIDIZATION BETWEEN ASIATIC AND NEW WORLD COTTONS *

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(With Plates IV and V)

INTRODUCTION

IT is a well known fact that cotton presents two taxonomically different groups, associated with differences in chromosome numbers. Cultivated cottons have been grouped as Asiatics or Old World cottons with 13 pairs of chromosomes and New World cottons with 26 pairs of chromosomes. As a rule, hybridization within one group leads to fertility and between the groups to sterility.

Recent cytological researches have brought to light some new facts concerning various wild species of cottons, and it has been shown [Skovsted, 1934] that New World cottons with $2n=52$ chromosomes are probably amphidiploids resulting from chromosome doubling in a hybrid between an Asiatic cotton with $2n=26$ and a wild American cotton with $2n=26$ chromosomes, though recently some doubts in this connection have been expressed by Gates [1938].

In Asiatic group, Surat cottons represent the high water level of quality cottons in India. The highest spinning value yet attained in any of the types of Indian cottons is 40's warp counts and strains spinning to this limit are in cultivation. To increase this spinning quality to a range higher than 40's is an important economic problem. There are no other types of Asiatic cottons available which can be utilized for crossing in order to increase the spinning value above this limit.

On the other hand, New World cottons including various American, Egyptian, and Sea-Island types which are reputed for long, silky and strong fibre (and consequently possessing very high spinning value), bigger bolls and greater vigour have been imported and efforts made to introduce them in general cultivation in various cotton growing tracts of India. But such attempts have in most cases met with indifferent success, especially with regard to longest stapled varieties, i.e. Sea-Island and Egyptian.

In view of this situation, it has often been suggested, 'why not cross American and Indian types and try to get a much better type for cultivation in India'. The idea of combining the good qualities of both these groups of cotton in a hybrid has always been a fascinating goal for many workers in cotton since long.

* This work has been recently financed by the Indian Central Cotton Committee at a cost of Rs. 14,014, spread over a period of five years commencing from 1 November 1938. The aim of the scheme is to combine the hardiness and adaptability of Asiatic cottons with the superior fibre qualities of the New World cottons.

PREVIOUS WORK

The recorded evidence of efforts at hybridization between Asiatic and New World cottons begins with Major Trevor Clarke's experimental work in the seventies of the nineteenth century. He failed in the attempt and, as quoted by Watt, is reported to have said, 'I strongly doubt the possibility of any cross between exotics and Indian sorts, and fear you will be disappointed in this respect when your plants come to perfection'. However, it is curious that Watt himself took it for granted that hybridization between these two groups was possible [Harland, 1928], and the natural evolution of some forms of cottons is attributed by him to this source. Mell [1897] reported complete failure in obtaining successful hybrids between American and Indian types. Webber [1905] was unable to cross 'Aiden' cotton (as quoted by him), which he classified as *G. herbaceum*, with varieties of either Sea-Island (*G. barbadense*) or Upland (*G. hirsutum*). Gammie appears to have been the first to obtain successful hybrids between these two groups, as reported by Watt [1907] and by himself [Gammie, 1905]. He produced hybrids between *G. hirsutum* × *G. obtusifolium*., *G. hirsutum* × *G. roseum* and *G. arboreum* × *G. hirsutum*, but nothing further is known about the results of his work.

The main credit must go to Zaitzev, who tackled the problem systematically and was able to get two hybrids between *G. herbaceum* × *G. hirsutum*. He crossed nearly one thousand flowers by his method of complete emasculation of removing the staminal sheath and obtained two hybrids thus giving 0.2 per cent success. He observed that only a few seeds were obtained per boll in case of crossed bolls. Five more hybrids of the same parentage were also observed by his assistants in the field as natural hybrids. As all his hybrids proved completely sterile, he abandoned the hope of getting anything further from them and finally regarded them as useless.

Accordingly, many workers endeavoured to get hybrids between these groups but were unsuccessful and, in consequence, it came to be generally believed that hybridization between the two groups was not possible or that hybrids, if produced, were sterile and of no further potential value. However, Vycotski [1930] reported upon the revival of such work on a large scale at Taskhent, and stated that the percentage of natural crossing between varieties differing in chromosome numbers was generally less than 0.003 per cent. Later, Kanash [1932] reported that he had been able to get 56 hybrids between various American and Indian types and had succeeded in inducing fertility in them by backcrossing. He observed that the percentage of successful hybrids in individual classes varies to a somewhat high degree, i.e., from 0.13 to 2.5 per cent; this variation being observed both between different crosses and in one and the same cross in different years. On the whole, success percentage varied from 0.011 to 2.25. He gives details of the phenomenon of occurrence of one seed in hybrid bolls as observed by him. Nakatomi [1931] got six hybrids between Asiatic and New World cottons. He crossed thousands of flowers, and his percentage success seems to be very low. Longley [1933] reported a hybrid between these two groups. Feng [1935] stated that he had obtained a few hybrids between *G. arboreum* and *G. nanking* with *G. barbadense* and *G. hirsutum*. He crossed 1,700 flowers and got seven hybrids, a percentage success of 0.4. He also observed the phenomenon of the occurrence

of one seed per boll in case of crossed bolls. Harland [1932, 1935] obtained two hybrids between *G. barbadense* and *G. arboreum* and by back-crossing to the higher chromosome parent, succeeded in getting fertile derivatives from the cross, introducing at the same time some of the genes from the Asiatic cotton into the other group. He crossed thousands of flowers and his success percentage seems to be very low. Szymanek [1932, 1936] reports to have got hybrids between these groups, but, from the complete fertility of the hybrids, the behaviour of the hybrid progeny and the results of cytological studies (cited by Skovsted), it seems that there was some doubt about the classification of the cottons he had under experimentation. Skovsted and Webber have done extensive scale hybridization between different groups of cotton. This work, together with cytological studies carried out by them, has thrown considerable light on the problem of the relationship between different species of cotton.

In India, Desai [1927] was the first worker to secure a hybrid between *G. hirsutum* × *G. herbaceum* and reported having obtained some successful first back crosses. Patel [1933] describes a hybrid obtained by him between *G. purpurascens* and *G. herbaceum*. He crossed only fifty flowers and secured one hybrid from one seed obtained from a single hybrid boll. The hybrids obtained by these two workers died on account of accidents and nothing further was achieved by them. Patel, G. B. (unpublished) produced a hybrid between *G. hirsutum* and *G. herbaceum* and Patel, P. L. (unpublished) also secured three hybrids of the same parentage at Broach. Few plants from backcrosses with *G. hirsutum* were also obtained and are at present growing at Broach. Ramanathan [1932] secured a hybrid between Karungani (*G. arboreum*) × Cambodia (*G. hirsutum*) which died later on due to stem weevil attack.

WORK AT SURAT

As a result of the impetus derived from the successful work of interspecific hybridization carried on in other crops and the successes obtained by Harland and Russian workers, research work in this direction was started in 1932 at Surat. Dr Burns, the then Director of Agriculture, Bombay Province, expressed his opinion that continued efforts should be made to obtain a fertile hybrid between Indian and American types of cotton in order to overcome the comparatively narrow limits set up by selection and crossing within either group. Preliminary information about the work was published by Thakar and Amin [1936] and Amin [1937], and the present paper is a comprehensive account of the whole investigation ever since it was started including the results previously published.

(a) Material

To begin with, work was started with a few Dharwar American and Punjab American types (*G. hirsutum*) grown on the station for the purpose, as it was considered that these types would be better suited to the climate than others. Later on, Sea-Island, Maarad (*G. barbadense*), and a few tree cottons (*G. religiosum* and *G. barbadense*) were added. The Asiatic types used were 1027 ALF and White Flower (*G. herbaceum*) and Gaorani. 6. and Red Arboreum (*G. arboreum*).

(b) Methods

In the beginning, the technique of hybridization was the one that is commonly followed, viz. emasculation by removing anthers with a pair of forceps and protecting the emasculated flower with a paper bag. Modifications of this technique were made in course of the work, and later on, the method advocated by Doak [1934] was adopted, i.e. the splitting of the staminal column with the finger nail and the removal of the entire corolla with the whole andræcium in a single piece. This method removes everything from the flower except the pistil which is thus exposed completely. It also ensures less chance of pollen remaining on the corolla and andræcium on account of chance breaking of anthers. The flowers were emasculated in the evening of the day preceding anthesis and enclosed in paper bags. Pollination was done next morning. This method however was found to work well with American types in which the flowers are bigger than in Indian types where the operation of the removal of the corolla by the finger nail proved to be liable to compress the delicate soft ovary or to rupture the thin style. Hence, with Indian cottons, emasculation was done by removing the corolla with a scalpel and the anthers by a pair of forceps. This operation was done early in the morning of the day of anthesis and is facilitated by the large size of the bud and by the fact that the anthers in these cottons do not burst so early as in New World cottons.

The F_1 hybrids are self-sterile and in order to utilize the large number of flowers opening on them, dusting of open flowers with the pollen of parents was done for backcrossing and the crossed flowers were left unprotected. A careful watch was kept on developing bolls which were protected against damage by boll worms with paper bags with their mouths tied with a piece of thread. The thread of the label, attached to the boll for identification was used for the purpose. This technique has been found to give satisfactory results. In addition, all flowers, other than pollinated ones, were removed from the parent plants. This operation prolonged flowering and facilitated the work of hybridization. The seedlings were first raised in pots under protection and then transplanted in the field.

(c) Results of experiments, 1932-1938

To begin with, methods advocated by Desai [1927] were given a trial, but without success. Desai [1927] obtained 12, 20 and 0.5 per cent success by painting the stigmas with citric acid solution, citric acid *plus* cane sugar solution and without any treatment of the stigma respectively. No success was however obtained by the author by the use of citric acid solution or cane sugar solution alone or in combination. Kanash [1932] and Webber [1936] also failed to get any results by painting stigma with various solutions and the latter thinks that the reported success might be due to the development of parthenocarpic capsules. The writer agrees with Webber as he found that parthenocarpy is pronounced in cottons and one gets bolls resembling empty shells with a few immature seeds, some of which even show slight immature lint developed on them particularly in those cases where only a few bolls are left developing on the plants.

Crossing and backcrossing work was undertaken on an extensive scale (Table I). The results obtained show that success can be had in hybridization between the two groups of Asiatic and New World cottons. The F_1 hybrids have been successfully raised as also the backcrosses to New World cottons resulting in plants with some characters inherited from either group together with induced fertility.

All the hybrids and backcross plants are being maintained as ratoons. With a view to further propagation and maintenance, vegetative propagation by layering, cuttings, and grafting has been tried. Layering in unsuccessful while cuttings give varying success from 10 to 25 per cent. Grafting is very successful and a number of grafts have been secured by simple approach and bottle grafting methods.

DISCUSSION

From the foregoing account of the work done at Surat and from reference to previous literature on the subject it can be said that:—

(1) It is possible to secure hybrids between the two groups of Asiatic and New World cottons differing in chromosome number.

(2) The fact that very few hybrids have been obtained by various workers, coupled with the similar experience at Surat, shows the difficulty involved but, as stated by Kanash, it can be safely concluded that to succeed in the attempt crossing must be resorted to on a large scale.

(3) All the F_1 hybrids reported above, have proved to be self-sterile. However, backcrossing to higher chromosome parent has been successful as shown by the experiments of Desai, Kanash, Harland, Nakatomi, Patel and the author at Surat. This is very encouraging as it indicates the way out of the impasse of sterility. So far, results of induced fertility by repeated backcrossing to higher chromosome parent have been reported only by Russian workers and by Harland.

(4) Percentage success in hybridization: It may be deduced from the previous literature as well as from the experience at Surat that success percentage is likely to vary from 0.01 to 2.5 in crosses between these two groups of Asiatic and New World cottons.

Thomson [1930] has summarized the literature as regards relative difference in success of reciprocal interspecific hybrids and concluded that more success is generally obtained when the species with the higher chromosome number is used as the female parent. In cottons, Zaitzev believed that better results could be obtained if the Asiatic types were used as the female parent. Kanash stated that reciprocal crossing is equally possible and successful. However, closer examination of his data indicates that the percentage success is greater when the species with the higher chromosome number is used as the female parent. Feng made a definite statement to the effect that whenever the female parent is the one with the higher number of chromosomes more success is obtained. The data presented here also confirm this conclusion.

In case of backcrossing a similar phenomenon is observed. Success has so far been obtained when the F_1 hybrid (which has higher chromosome number) has been used as the female parent. Again, of the two species entering into the cross, backcrosses are successful with higher chromosome parent as the

TABLE I
Results of hybridization between Asiatic and New World cottons and the F_1 hybrids backcrossed to the parents

Year	Parents	No. of flowers pollinated	No. of Bolls set	No. of seeds obtained	No. of hybrids obtained	Success per cent	Remarks
	<i>Direct Crosses</i>						
1932-33	$\frac{\sigma}{\sigma}$ <i>G. hirsutum</i> \times <i>G. herbaceum</i>	432	Desai's methods were tried
1933-34	<i>G. hirsutum</i> \times <i>G. herbaceum</i>	1,924	
1934-35	<i>G. hirsutum</i> \times <i>G. herbaceum</i>	1,607					
	Dharwar American \times 1027 ALF		7	10	6	0.43	3 seeds failed to germinate. One hybrid died later.
	Punjab American \times 1027 ALF				1		
	<i>G. hirsutum</i> \times <i>G. arboreum</i>						
	Cambodia \times Red Arboreum	742	2	4	4	0.53	Two hybrids died later.
	<i>G. herbaceum</i> \times <i>G. barbadense</i>	1,683					
	1027 ALF \times Exotic 1				1		
	<i>G. herbaceum</i> \times <i>G. religiosum</i>		2	5		0.11	3 plants turned out to be like the mother parent
1935-36	1027 ALF \times Exotic 2				1		
	<i>G. hirsutum</i> \times <i>G. herbaceum</i>						
	Dharwar American \times White Flower	1,244			2		
	Cambodia \times 1027 ALF		21	22	1	0.64	14 seeds failed to germinate
	Punjab American \times 1027 ALF				5		
	<i>G. hirsutum</i> \times <i>G. arboreum</i>						
1936-37	Dharwar American \times Gacranl 6	889	4	4	2	0.22	Two seeds failed to germinate
	<i>G. barbadense</i> \times <i>G. herbaceum</i>						
	Maarad \times 1027 ALF	792	2	3	3	0.37	
		23	...	Total F_1 hybrids on hand

TABLE 1—*contd.*

Year	Parents	No. of flowers pollinated	No. of bolls set	No. of seeds obtained	No. of hybrids obtained	Success per cent	Remarks
<i>Backcrosses</i>							
1935-36	10 F ₁ hybrids of 1934-35 × Respective New World parents	15,000	3	3	All failed to germinate
	10 F ₁ hybrids of 1934-35 × Respective Asiatic parents	2,000	
	New World types × Hybrid pollen	6,716	
	Asiatic types × Hybrid pollen	500	
1936-37	20 F ₁ hybrids of 1934-35 and 1935-36 × Respective New World parents	100,000	38	41	20	...	21 seeds failed to germinate. One plant died later, thus 19 backcross plants on hand
	New World types × Hybrid pollen	2,000	
1937-38	Asiatic types × Hybrid pollen	2,000	
	23 F ₁ hybrids of 1934-35, 1935-36 and 1936-37 × Respective New World parents	196,000	47	51	15	...	36 seeds failed to germinate. 2 were damaged by insects and 5 died later, thus 8 backcross plants on hand
Percentage of 27 successful backcross plants							
1	<i>(G. hirsutum × G. herbaceum) F₁ × G. hirsutum</i>						
	(a) (Dharwar American × 1027 ALF) F ₁ × Dharwar Ameri-can	...	21				
	(b) (Cambodia × 1027 ALF) F ₁ × Cambodia	...	2				
2	<i>(G. hirsutum × G. arboreum) F₁ × G. hirsutum</i>						
	(a) (Dharwar American × Gaorani 6) F ₁ × Dharwar Ameri-can	...	2				
	(b) (Cambodia × Red Arboreum) F ₁ × Cambodia	...	2				

Fig. 1. *G. hirsutum* × *G. arboreum* F₁



Fig. 2. Characters of the hybrid as compared to parents



pollen parent. Harland who was foremost in trying backcrossing on a large scale in interspecific hybrids, has shown that success is only obtained when pollen from a parent with higher number of chromosomes is used on to the flowers of the hybrid plants. Nakatomi and Kanash have obtained success in backcrossing when a male parent with higher number of chromosomes was used. It may be noted that in backcrossing, no success has yet been reported when Asiatics, or the species with the lesser number of chromosomes, is used as one of the parents.

(5) Size of bolls and number of seeds per boll in crosses : The peculiar phenomenon of the occurrence of only a few seeds per boll in case of crossed bolls in species crosses has been observed by various workers. Thirty-two seeds from twenty-one crossed bolls were obtained, the average number of seeds per boll coming to 1.52, thus confirming the observations of the various workers. In backcrossing, the formation of bolls on the F_1 hybrids shows a similar phenomenon. Only one seed per boll is the rule with few exceptions. The total number of bolls obtained from the F_1 hybrids in the three seasons was 88 giving 95 seeds.

(6) General characters of the F_1 hybrids :—

(a) *Hybrid vigour* : All the F_1 hybrids showed very marked hybrid vigour. They were more than three to four times the size of the parents (Plate IV).

(b) *Other characters* : In many characters, these hybrids are generally intermediate (Plate IV). The hairiness, shape of leaf, colour of petal, colour of pollen, etc., are all intermediate. The petal spot of Indian cottons is dominant but the intensity varies in different hybrids. There is a peculiar phenomenon in that the presence of various grades of spot, or even its complete absence, occurred on different branches of the same hybrid plant. Zaitzev noted a similar phenomenon in his hybrids. These variations of the different grades of spots, or their complete absence on different branches of the same plant, are such that by vegetative propagation of such branches plants showing these differences were raised. In case of *Arboreum* crosses, the anthocyanin pigmentation on plant body, leaf and flower is reduced. The flower colour is in accordance with Harland's description, viz. 'yellow with a red flush at the petal edges. Spot intermediate between the parents'.

(c) *Sterility* : As mentioned before all the hybrids proved completely self-sterile, except in so far as a few seeds could be procured by backcrossing with New World parents.

(7) Inducing fertility by backcrossing on F_1 hybrids : During the last three seasons, out of twenty-three F_1 hybrids, thirteen have set bolls on backcrossing with the New World cottons as pollen parents, and, as a result, twenty-seven backcrossed plants have been raised therefrom. The setting of bolls on backcrossing has been rare, but it is worth noting that one hybrid (Dharwar American \times 1027 ALF) F_1 —Hybrid No. 2-4 (which on cytological examination has been found to be a tetraploid)* has set a significantly large number of bolls consistently during the last three seasons, the success percentage being

0.16, 0.49, and 0.21 for each of these seasons respectively. In fact, out of the twenty-seven backcross plants at present on hand, fifteen are derived from the tetraploid hybrid.

(8) Fertility in first backcross population : Of the twenty-seven plants grown, fifteen have set bolls on second backcrossing or selfing. Special mention may be made of a fully fertile plant No. 22 (Plate V). It has a red plant body and red flower colour with petal spot inherited from Asiatic parent. The character behaves as a simple dominant one as observed in the selfed and backcrossed progeny plants, with varying intensity of its expression. The progeny of first backcross plant No. 22 is fully fertile (Plate V). Normal setting of bolls also occurs by using this plant as a male or female parent in crosses with New World cottons. The fertility observed in most of the first backcrosses is encouraging in comparison with the results obtained by Harland who got full fertility after four backcrossings.

(9) There are many known physical and chemical treatments for getting fertility in interspecific hybrids by inducing doubling of chromosomes. Notable among these are, physical injury by wounding, ringing or callus formation, heat and cold treatment, X-ray applications, the use of chemicals, e.g. anaesthetics and narcotics such as chloroform and colchicine. Of these methods, wounding and ringing have been practised but without success. Trials were made at callus formation and though such is possible in cottons, sprouting of shoots from the callus did not take place. Anaesthetic like chloroform was also used but without success. The use of colchicine as an agent for inducing doubling of chromosomes appears to be promising, and experiments with it on cottons are being conducted by the author and results are awaited. If some such method proves successful, the question of inducing polyploids in cottons will be an easy task opening up a wide field for further research.

SUMMARY

After summarizing the general position of interspecific hybridization in cottons, with references to the literature on the subject, details of similar work carried out at Surat for six seasons from 1932-38 are given.

In all, 23 F_1 hybrids between Asiatic and New World cottons have been grown. Backcrossing to New World cottons has proved successful and, as a result, twenty-seven first backcrosses have been obtained, many of which have also shown fertility through second backcrossing and selfing.

Data regarding the percentage success in hybridization, hybrid boll characters, general characters of the hybrids including sterility and fertility are discussed.

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* 21 F_1 hybrids have been kindly examined cytologically by the Geneticist and Botanist, Institute of Plant Industry, Indore. All are triploids ($2n=39$) except two of which one is tetraploid ($2n=52$) and one pentaploid ($2n=65$).



FIG. 2. One of the selfed progeny plants of B C No. 22



FIG. 1. Fertile first back cross progeny plant, B C No. 22
(Cambodia \times Red Arboreum) $F_1 \times$ Cambodia

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GENETICAL STUDIES IN *COFFEA ARABICA* L.

A PRELIMINARY STUDY WITH YOUNG LEAF COLOUR AND RIPE PERICARP COLOUR

BY

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INTRODUCTION AND HISTORICAL

THE colour of young leaves and the colour of pericarp of ripe fruits of *Coffea arabica* L. show well-marked differences. Sometimes varieties and strains have been distinguished by these characters. Taschdjian [1932] reports that *Café Bourbon* is 'to be distinguished by the light green colour of the young leaves in contrast to the reddish to bronze of those of *Café Nacional*.' Krug [1935] mentions, "*Como é do conhecimento dos que trabalham com a Botânica do cafeeiro, constata-se as seguintes cores nos brotos do cafeeiro: a) verde (com diversas tonalidades desde verde quasi branco a um verde escuro), que caracteriza os representantes typicos das variedades Mokka, Laurina, Murta e Bourbon; b) bronze (de bronze claro até muito escuro) encontrado, de preferencia, nas variedades arabica typica (Nacional incluindo a forma aqui chamada de café Sumatra) e amarella (Amarello de Botucatu) e, finalmente, c) purpureo caracteristica da variedade purpurascens 'Café Roxo'.*" About the colour of pericarp of ripe fruits Krug [1936] mentions, '*Com relação com os frutos presentemente nada podemos adeantar como, e sabido, a cor vermelha é a mais commum sendo a amarella caracteristica da var Amarella. (Amarello de Botucatu) é das variedades (?) Maragogipe e Bourbon Amarelos. A variedade Purpurascens caracteriza-se por frutos tambem arroxeados.*'

Recently, genetical work has been carried on using the colour of young leaves and the colour of pericarp of ripe fruits as characters for the study of inheritance in coffee. Krug [1936] mentions that controlled pollination has shewn a number of the plants selfed to be heterozygous. Bronze colour of the young leaves was dominant (though incompletely) to green; both these were dominant to purple. Stoffels [1936] describes the 'behaviour of two groups of *arabica* coffee, one with brown leafed terminal shoots BB and the other with green, BV..... The effect of the environment on variation was very marked but the BB group proved much more adaptable than the BV group and less susceptible to dieback and black tip and to over production.

'Most of the BB strains tested were homozygous with regard to the colour of their terminal shoots but in some cases, although the numbers were too small for certainty, there were indications of a monohybrid ratio for the characters BB and BV. Which character was dominant depended on the mother plant.' He reports again, 'It was observed that the progeny of the so-called local varieties were some (BB) and some (BV) while those of the introduced coffees were all (BB).' [Stoffels, 1937].

Dealing with fruit colour Taschdjian [1932] mentions, 'The results of the cross *D'Utra* × *Amarello* indicate that the yellow colour is dominant to red.'

Coffea arabica L. is not the only species in which the pigmentation of the leaves has been made use of for genetical study. Lewis and Crane [1938] have investigated the anthocyanin pigmentation of leaves and shoots of apples. They mention, 'The anthocyanin coloration is a convenient character for genetic study, as it develops in the early seedling stage and the plants can be scored and the majority discarded while quite small, thus obviating the labour and expense of growing trees to maturity.' This fact may also be made use of with coffee.

MATERIAL AND METHODS

A large number of varieties and strains of *Coffea arabica* L. has been collected and plants have been raised from these at the Government Coffee Experiment Station, Balehonnur. Several mother plants were selected from among these. For progeny tests seeds were obtained from the mother plants by controlled self-pollination. The seedlings raised have been planted in the field. The progeny of each mother plant is numbered. Thus, family 288 is the progeny of the mother plant S. 26. The mother plants and selections in their progeny have been crossed with pollen from Kent and Coorg strains of *arabica* coffee. These several families as well as the different strains and varieties formed the material for observation.

The characters studied were colour of young leaves and colour of pericarp of ripe fruits. The colour of young leaves that have just unfolded from the leaf bud was studied as, during the attainment of maturity the different colours change to the normal green colour. In the text wherever the colour of leaves is mentioned it refers to the colour of young leaves that have just unfolded from the leaf bud.

Three groups have been distinguished for the colour of young leaves viz. copper, brown, and light green. These appear to correspond with purple, bronze, and green colours of new leaves in Fig. 3 of Krug [1936].

While in the light green colour group the colour of leaves is uniform the same cannot be said of leaves in the copper and brown groups. Within the copper and brown classes the amount of coloration varies from only a slight tinge of copper red or brown to intense copper red or brown. There is no difficulty, however, in distinguishing the two groups. In this paper only the three broad classes—copper, brown, and light green—have been distinguished.

Pericarp colour of ripe fruits falls into two groups, viz. red and golden-yellow. These appear to correspond with red and yellow colours of fruits in Fig. 4 of Krug [1936].

DESCRIPTION

Kent's strain of *arabica* coffee has young leaves with copper colour. A major portion of cultivated coffee on the Station—Coorg strain—has copper leaves, a minority having brown leaves. Both the strains produce fruits with red pericarp. Golden Drop coffee—known as Amarella in Brazil [Cramer, 1913]—is characterized by light green leaves and fruits with golden-yellow

pericarp. Interspersed among the cultivated coffee wherein supply-planting has been carried on for some time, are a number of plants that have light green leaves and golden-yellow pericarp. All the observations I have made so far indicate that plants with copper and brown leaves have red pericarp, and, plants with light green leaves have golden-yellow pericarp.

TABLE I

Segregation in 1st and 2nd generation progeny of mother plants

Mother plant	Progeny Family No.	Colour of leaves of mother plant	Colour of pericarp of mother plant	Colour of leaves of progeny No. of plants with			Colour of pericarp of progeny No. of plants with	
				Copper	Brown	Light green	Red	Golden-yellow
S 26	288	Brown	Red	24	34	13	58	13
S 26	526		"	22	33	14
288-5	498		"	21	4	14
288-20	467	"	"	12	33	30	45	30
288-20	527		"	22	36	27
288-40	497	Copper	"	16	19	8
288-22	496		"	8	7	0
288-22	529		"	18	15	0
288-23	468	"	"	179	59	0	238	0
288-70	495	"	"	50	23	0
288-53	466	Light green	Golden-yellow	0	0	12
288-53	500		"	0	0	16
288-53	531		"	0	0	57
S 32	350	Brown	Red	15	18	14	33	14
S 48	360	"	"	7	23	0	30	0
S 13	356	Light green	Golden-yellow	0	7	24	7	24
S 63	375	"	"	0	72	32	72	32
S 44	353	Copper	Red	237	64	0	301	0
353-53	535	"	"	26	0	0
S 59	371	"	"	43	23	0	66	0
Kent	446	"	"	100	0	0	100	...
446-11	578	"	"	128	0	0
Kent	450	"	"	100	0	0	100	0
450-26	579	"	"	64	0	0
Kent	451	"	"	100	0	0	100	0

N.B.—The blanks in the last two columns indicate that the plants in those families have not fruited.

First generation progeny of three Kent and second generation progeny of two Kent plants have bred true for copper leaves. S 44 and S 59 have segregated in their first generation wherein, the plants have copper and brown

leaves. Second generation progeny of S 44 obtained from selfing one plant with copper leaves in the first generation, have only copper leaves.

In its first generation S 26 has segregated : there are plants with copper, brown, and light green leaves. Second generation progeny from selfing three plants with copper leaves in the first generation, have copper and brown leaves ; second generation progeny from selfing three plants with brown leaves in the first generation, have copper, brown, and light green leaves ; while, three families from selfing one plant with light green leaves in the first generation, have all light green leaves.

S 32 and S 48 have segregated in their first generation : progeny of S 32 have copper, brown, and light green leaves ; and, the progeny of S 48 have copper and brown leaves only.

The first generation progeny of S 13 and S 63 have brown and light green leaves.

TABLE II

Results of crossing mother plants and their progeny with Kent and Coorg strains

Parents	Family No.	Colour of leaves of parents		Colour of pericarp of parents		Colour of leaves of F ₁ Hybrids. No. of plants with			Colour of pericarp of F ₁ Hybrids. No. of plants with	
		Female parent	Male parent	Female parent	Male parent	Copper	Brown	Light green	Red	Golden-yellow
S 44 ♀ × Kent ♂ .	351	Copper	Copper	Red	Red	112	0	0	112	0
S 44 ♀ × Coorg ♂ .	352	"	"	"	"	79	0	0	79	0
S 26 ♀ × Kent ♂ .	327	Brown	"	"	"	145	133	12	278	*2
S 26 ♀ × 286-14 (Kent)	490	"	"	"	"	72	32	1	104	...
S 26 ♀ × Coorg .	516	"	"	"	"	46	22	0
238-20 ♀ × 286-14 ♂ (Kent)	493	"	"	"	"	32	11	0	43	0
238-20 ♀ × Coorg ♂ .	518	"	"	"	"	125	72	0
238-53 ♀ × 446-11 ♂ (Kent)	569	Light green	"	Golden yellow	"	110	29	0
446-11 ♀ × 238-53 ♂ (Kent)	571	Copper	Light green	Red	Golden yellow	96	34	0
S 48 ♀ × Kent ♂ .	395	Brown	Copper	"	Red	88	14	0	102	0

N.B.—The blanks in the last two columns indicate that the plants in the families have not fruited.

* Of the 12 plants with light green leaves in the family only two have fruited.

S 44 has been crossed with pollen from a Kent and a Coorg plant. The F₁ hybrids have all copper leaves.

On crossing S 26 with pollen from two Kent plants the F₁ hybrids obtained have copper, brown, and light green leaves ; while, crossing S 26 with pollen from a Coorg plant the F₁ hybrids have copper and brown leaves only. One plant with brown leaves in the first generation progeny of S 26 has been crossed with pollen from a Kent and a Coorg plant. The F₁ progeny have copper and brown leaves.

The F_1 plants from the cross S 48 \times Kent have copper and brown leaves.

One plant with light green leaves in the first generation progeny of S 26 has been crossed reciprocally with a Kent. In both the crosses the F_1 hybrids have copper and brown leaves.

TABLE III

Reciprocal intercrosses in the first generation progeny of S 26

Parents	Family No.	Colour of leaves of parents		Colour of leaves of F_1 hybrids No. of plants with		
		Female parent	Male parent	Copper	Brown	Light green
288-20 ♀ \times 288-53 ♂	562	Brown	Light green	14	16	38
288-53 ♀ \times 288-20 ♂	566	Light green	Brown	12	53	96
288-23 ♀ \times 288-53 ♂	564	Copper	Light green	37	37	15
288-53 ♀ \times 288-23 ♂	567	Light green	Copper	92	70	6

N.B.—The plants in the families have not fruited.

When plants with brown and light green leaves in the first generation progeny of S 26 are reciprocally crossed the F_1 progeny have copper, brown, and light green leaves. The plants with light green leaves are in excess. On crossing reciprocally plants with copper and light green leaves in the first generation progeny of S 26, the F_1 hybrids have copper, brown, and light green leaves. The plants with light green leaves are in a minority.

DISCUSSION

Krug [1935] reports, '*Constatarem-se as seguintes condicoes de dominancia : Bronze e, incompletamente dominante sobre verde, sobre os hybridos (F_1) de coloracao intermedia. Quanto ao purpureo, constatou-se que esta coloracao e recessiva tanto em relacao com o verde, como com o bronze. Somente a segunda geracao filial (F_2) e os 'back crosses' poderao revelar o numero de gens que condicionam a cor nas folhas novas.*' In a later publication he mentions [Krug, 1936], '*Nas variedades de cafe cultivadas sao duas as cores predominantes das folhas novas : bronze e verde ; trata-se, provavelmente, de um unico par de factores (Br-Br, br-br) apresentando o heterozygo to Br-br. dominancia incompleta de Br, pois e bronze claro. A variedade Purpurascens possui folhas novas purpureas ; hybridos desta variedade com cafeiros de folhas novas verdes ou bronzeadas apresentam dominancia completa destas duas cores sobre o purpurascens ; nada podemos, no emtanto, adiantar sobre si todos estes factores sao allelomorphos ou si o Br e o br sao epistaticos sobre um outro factor independente que determina a cor purpures*'.

Stoffels [1936] reports that Most of the BB strains tested were homozygous with regard to the colour of their terminal shoots but in some cases, although the numbers were too small for certainty, there were indications of a monohybrid ratio for the characters BB and BV. Which character was

dominant depended on the mother plant.' Again, 'Eight introduced varieties and 26 out of 33 lines of local varieties proved to be homozygous with regard to this colour character as well as to other morphological characters associated with leaves, stems and internodes. Seven lines of Mibirizi, on the other hand, showed a 3 : 1 segregation for leaf colour in the F_2 progeny, but whether BB or BV was dominant depended on whether the mother tree was BB or BV.' [Stoffels, 1936].

(a) *Leaf colour : segregation in first and second generation progeny*

(1) *Copper colour*.—Kent's strain appears to be homozygous for copper leaves: the first and second generation progeny have bred true for copper leaves. S 44, S 59, and plants with copper leaves in the first generation progeny of S 26, suggest the dominance of copper colour over brown in their progeny. In family 468 the segregation into copper and brown classes is in accordance with a 3 : 1 ratio; whereas, in families 353, 495, and 371 the segregation is not inconsistent with a 3 : 1 ratio. The remaining two families—496 and 529—are very small. Thus, there appears to be a single factor difference between copper and brown classes.

(2) *Brown colour*.—S 26, S 32, and plants with brown leaves in the first generation progeny of S 26, segregate in their progeny which have copper, brown, and light green leaves. The segregation of S 26 is not inconsistent with a 1 : 2 : 1 ratio between copper, brown, and light green classes; whereas, with the remaining families there is no such clear cut segregation. The progeny of S 48 have only copper and brown leaves; but, the family is too small to be relied upon. The data so far obtained indicate that plants with brown leaves are heterozygous.

(3) *Light green leaves*.—S 13 and S 63 segregate differently in their progeny. The segregation into brown and light green classes in the progeny of S 63 is not inconsistent with a monohybrid ratio. However, the presence of excess of plants in the brown group in the progeny of S 63 is reversed in the progeny of S 13. S 13 and S 63 are two examples of the dominance of light green over brown leaves.

The plant with light green leaves in the first generation progeny of S 26 appears to be homozygous for leaf colour as its progeny breed true for the character.

(b) *Leaf colour in crosses*

(1) *Copper colour*.—The F_1 hybrids of the crosses S 44 \times Kent and Coorg have all copper leaves. The F_1 progeny of the reciprocal crosses of a Kent and a light green leaved plant in the first generation progeny of S 26, have copper and brown leaves the former being in excess. These reciprocal crosses are of particular interest: both the parent plants are homozygous for leaf colour. In the F_1 progeny the copper colour of Kent dominates over the light green colour in producing an excess of plants with copper leaves; and, also, the copper colour of Kent combines with the light green colour in producing a minority of plants with brown colour.

The F_1 hybrids of the reciprocal crosses between a plant with copper leaves and a plant with light green leaves in the first generation progeny of S 26 have copper, brown, and light green leaves, plants with copper leaves

being in excess. The copper leaved parent is not pure but, contains factors for brown colour and, hence, the presence of plants with green colour in the crosses is not unexpected. However, the presence of plants with copper leaves in excess in the crosses is suggestive of the dominance of copper over light green.

(2) *Brown colour*.—In the F_1 hybrids of the crosses between plants with brown and copper leaves, plants with copper leaves are in excess. In two of the crosses between S 26 and Kent, there are present a few plants with light green leaves. In these crosses there is a suggestion of the dominance of copper over brown.

The reciprocal crosses between 288-20 and 288-53 have F_1 hybrids with copper, brown, and light green leaves, the last one being in excess. As 288-20 produces a great percentage of plants with light green leaves in its progeny the presence of an excess of plants with light green leaves in the reciprocal crosses between 288-20 and 288-53 is according to expectation.

(c) *Colour of pericarp of ripe fruits.*

Pericarp colour appears to be linked up with leaf colour: all the observations so far made show that red pericarp is linked up with copper and brown leaves, and golden-yellow pericarp is linked up with light green leaves. Therefore, the segregation of pericarp colour follows closely the segregation of leaf colour in the several progeny.

CONCLUSION

Tests on mixing solutions of intense red and light green colours have resulted in brown colour. On crossing plants that are homozygous for copper and light green leaves, the F_1 hybrids have some plants with brown leaves. Further, plants with brown leaves segregate in their selfed progeny into copper, brown, and light green classes. These facts indicate that brown leaves might be in the nature of a blend between copper and light green leaves.

The segregation in the progeny of plants with copper leaves into copper and brown classes is suggestive of a single factor difference between the two classes. Likewise, the segregation in the progeny of S 26 into copper, brown, and light green classes, and the segregation into brown and light green classes in the progeny of S 63, appear to be not inconsistent with a monohybrid ratio. Further, there are indications that copper leaves dominate over light green leaves and brown leaves; and, light green leaves dominate over brown leaves.

Plants have been met with that are homozygous for copper and light green leaves. All the plants with brown leaves appear to be heterozygous.

The pericarp colour of ripe fruits appears to be linked up with leaf colour: red pericarp being characteristic of plants with copper and brown leaves; and, golden-yellow pericarp characterizes plants with light green leaves.

SUMMARY

(1) Colour of young leaves of *Coffea arabica* L. that have just unfolded from the leaf-bud, as well as, the colour of pericarp of ripe fruits show well marked variations. The young leaves have copper, brown, or light green colours; the pericarp has either red or golden-yellow colour. These characters have been made use of for genetical study.

(2) Colour of pericarp of ripe fruits appears to be linked with leaf colour : plants with copper and brown leaves have red pericarp ; and, plants with light green leaves have golden-yellow pericarp.

(3) Plants homozygous for copper and light green leaves have been met with ; whereas, all the plants with brown leaves appear to be heterozygous.

(4) Copper colour of leaves combining with light green colour in the crosses between the two have produced some plants with brown leaves.

(5) Segregation in the progeny of plants with (a) copper and, (b) light green leaves into (a) copper and brown classes, and, (b) brown and light green classes respectively, seems to be not inconsistent with a monohybrid ratio. Segregation into copper, brown, and light green classes in the progeny of plants with brown leaves does not appear to be so clear cut.

(6) Copper leaves appear to dominate over light green and brown leaves ; and, light green leaves appear to dominate over brown leaves.

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INSECT POLLINATORS OF *TORIA* (*BRASSICA NAPUS*
LINN., VAR. *DICHOTOMA* PRAIN), AND *SARSON*
(*B. CAMPESTRIS* LINN., VAR. *SARSON* PRAIN)
AT LYALLPUR

BY

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INTRODUCTION

A number of economic plants (e.g. red clover, Cucurbitaceæ, some of the Brassicæ, apple, etc.) depend entirely upon insects for pollination [Waite, 1895, '99; Treherne 1923; Williams, 1925]. An increase in the population of such insects results in a material increase in the yield of the crop [Alderman, 1918; Auchter, 1922; Farrar, 1931].

In India this fruitful line of investigation has remained practically untouched. Apart from Burkill's random notes [1906-09, 1911], Ali Mohd. *et al's* [1931] and Ali Mohd.'s [1935] articles are the only contributions to the subject. The reason for this neglect is not far to seek. Economic Entomologists have been, and are still, busy combating injurious insects to realize the full significance of this branch of their subject, the importance of which to a pre-eminently Agricultural country like India, cannot be over-emphasized. It is, therefore, reasonable to maintain that a more intimate and comprehensive knowledge of insect visitors to the flowers of our plants of economic importance is long overdue.

OBJECTS OF THE PRESENT INVESTIGATION

Apropos of the record low yield of *toria* during 1928-29 in the Punjab Afzal Husain [1930] made the following observation: 'The last *toria* crop failed, and it was noticed that although the number of pods per plant was fairly large, the number of seeds per pod was very little, which shows defective pollination. As this crop is pollinated by insects, it seems likely that on account of certain unknown causes insects pollinating *toria* were not present in large numbers. It is a well known fact that insect pollinators play a most important part in the economy of nature and the question is so important that it cannot be ignored much longer.'

The work on insect visitors to *toria* and *sarson* flowers was, therefore, begun in January, 1930, at Lyallpur with a view to determine: (1) the species of insect visitors and their importance as pollinating agents, and (2) the relative significance of the important pollinators. This information was considered an essential preliminary for the elucidation of those 'unknown causes' which retard insect activity.

THE SPECIES OF INSECT VISITORS TO *TORIA* AND *SARSON* FLOWERS AND THEIR IMPORTANCE AS POLLINATORS.

Method of study.—The insect visitors to *toria* and *sarson* flowers were collected* for about 131 days i.e. from November to February, 1930 and 1931, in the Botanical Experimental Farm, Lyallpur. Collections were started almost immediately after the flowers appeared and were usually made between 12 noon to 3 P.M.—the period of greatest insect activity—for an average of 2·17 hours a day.

Weather conditions during collection period.—Excepting eight days†, when the prevailing weather conditions were not favourable, the remaining 123 days, particularly during the collection period, had ideal conditions for insect flight and activity. Table I gives the meteorological data which was found favourable for insect flight during *toria* and *sarson* seasons in 1930 and 1931.

Insect visitors.—The collections made include 105 different species representing 55 families of 9 orders of the Class Insecta. The importance of these insects as pollinating agents is discussed below.

ORDER HYMENOPTERA

Fam. APIDAE.—*Apis florea* Fab. is the only representative of this family secured from *toria* and *sarson* flowers. Table II gives the numbers collected from the two crops during the specified weeks.

It is seen from the above table that *Apis florea* Fab. is slightly more abundant in *sarson* than *toria*. It begins to desert this crop for other flowers towards the end of February.

The nesting habits of *Apis florea* Fab. are described by Ghosh [1915].

This bee works zealously and enthusiastically 'during bright, sunny, warm weather', when there is slight or no wind. During inclement weather it does not leave its hive. When overtaken by bad weather during work it immediately suspends its activities and clings to the flower. In this condition it allows itself to be picked up by hand without stinging.

Apis florea Fab. works in a purposeful manner. Table III gives the number of flowers visited per minute.

Thus during favourable weather *Apis florea*, Fab., on an average, visits six flowers per minute.

Its *modus operandi* on flowers is ideal for cross-pollination. Alighting on a flower it sends its proboscis down immediately to a nectary and in consequence the head comes in contact with anthers and gets heavily dusted with pollen. The next nectary is reached with the body usually lying across the anthers. Much more frequently, however, the bee lies across the apex of the stigma to lap up nectar: it is this posture that results in pollination.

* The collections were made by Messrs. K. G. Bhandari, B. Singh, R. Takoo, N. Kishore, the late S. Datt, Sana Ullah and the author.

† 12th and 24th January, 1930.

27th February, 1930.

24th November, 1930.

7th and 10th February, 1931.

TABLE I
Showing meteorological data favourable for insect flight during toria and sarson seasons in 1930 and 1931 respectively

Month and year	Flowering period of	Dates	Collection made for		Mean maximum temperature (F)	Mean minimum temperature (F)	Mean humidity per cent	Amount of sun-shine		Amount of rain-fall (inches)	Wind velocity	
			Hrs.	Mts.				Hrs.	Mts.		Maximum, Miles	Minimum per hour
November 1930	<i>toria</i>	19-25	15	..	77.85	48.28	66.71	50	30	..	2	1
December 1930	<i>toria</i>	1-7	13	..	78.14	45.28	60.71	44	30	..	2	1
January 1930	<i>sarson</i>	13-19	16	30	62.85	36.28	81.287	3	1
February 1930	<i>sarson</i>	13-19	6	..	78.00	47.43	72.42	2	1
November 1931	<i>toria</i>	11-17	12	..	82.42	49.00	53.42	62	25	..	3	1
December 1931	<i>toria</i>	7-13	14	..	73.28	43.28	73.28	53	15	..	3	1
January 1931	<i>sarson</i>	24-30	12	30	66.28	38.14	72.00	47	48	..	3	1
February 1931	<i>sarson</i>	13-19	10	..	67.42	41.85	74.14	44	42	..	3	1

TABLE II

Showing numbers of *Apis florea* Fab. collected from toria and sarson during 1930 and 1931

Name	1930		1931	
	<i>toria</i> (November 19-25 December 1-7)	<i>sarson</i> (January 13-19 February 13-19)	<i>toria</i> (November 11-17 December 7-13)	<i>sarson</i> (January 24-30 February 13-19)
<i>Apis florea</i> Fab.	62	188	216	370

TABLE III

Showing number of sarson flowers visited per minute by *Apis florea* Fab.

Date	Time of observation		Duration of observa- tion (minutes)	Total number of flowers visited	Number of flowers visited in one minute
	From P.M.	To P.M.			
19-1-30	12.1	12.7	6	30	5
19-1-30	1.5	1.6	1	8	8
19-1-30	2.29	2.32	3	22	7.3
8-2-30	2.34	2.39	5	37	7.4
14-2-30	2.42	2.47	5	25	5
26-1-31	2.52	2.54	2	10	5
9-2-31	2.53	2.55	2	16	8
12-2-31	2.58	3.9	11	45	4
14-2-31	2.41	2.49	8	41	5.1

Fam. ANDRENIDÆ.—This family was strongly represented. Table IV gives the names and numbers of the members of this family collected from *toria* and *sarson* during the specified weeks.

It is seen from Table IV that *Andrena ilerda* Cam. and *Halictus* sp. are more abundant in *sarson* and *toria* crops than any other species of *Andrenidæ*. Both these solitary bees are exceedingly industrious and work in a purposeful manner. Because of these qualities and their numbers and body structure these bees are among the most important pollinators of flowers. Their *modus operandi* on flowers is identical with that of *Apis florea* Fab.

At Lyallpur *Andrena ilerda* Cam. begins to visit *toria* flowers in the second week of November. It reaches its maximum numbers in December : in fact it is during this month that it predominates over all other insect visitors. It constructs its nest in and around *toria* fields. For this purpose it makes a 22 in.—25 in. long tunnel with a number of side tunnels along its course.

In the case of an allied species each side of tunnel is said to terminate in a cell in which balls of 'bee-bread' (for egg-laying) are stored.

TABLE IV

Showing names and numbers of *Andrenidae* collected from toria and sarson during 1930 and 1931

Name	1930		1931	
	<i>toria</i> (November 19-25 December 1-7)	<i>sarson</i> (January 13-19 February 13-19)	<i>toria</i> (November 11-17 December 7-13)	<i>sarson</i> (January 24-30 February 13-19)
<i>Andrena ilerda</i> Cam.	299	25	384	23
<i>Halictus</i> sp.	22	51	112	46
<i>A. leena</i> Cam.	8	3	..	10
<i>A. satellita</i> Nurse	..	3	..	17
<i>Halictus salsettensis</i> Ckll.	..	2	..	14
<i>Andrena</i> sp.	1	2	2	2
<i>Andrena</i> sp.	3	1	2	2
<i>A. ephippium</i> Spin. var. <i>dilecta</i> Mocs.	..	2
<i>A. fulvicrus</i> Kirby	2

Six observations were made to study the duration of a pollen-collecting trip. The method adopted was to keep a watch over the burrow from which an *Andrena ilerda* Cam. was seen coming out. (In all observations the burrow entrance was blocked with a clod which was removed when the insect returned).

It will be observed from Table V that the first visit of *Andrena ilerda* Cam. (apart from the first forced return in observation II) lasted for 3 to 4½ hours. The material collected during this trip required 7-10 minutes to be unloaded. The second visit lasted for about 1½ to 2½ hours, the insect taking 2-4 minutes to unload. The third visit was of the shortest duration for it lasted only from 19 minutes to 1½ hours. For pollination the first visit, and to a lesser degree the second visit, are most important, because 'an examination of the anthers of the freshly opened flowers at about 3 P.M. showed them to be absolutely devoid of pollen grains which are evidently carried away by their insect visitors' [Ali Mohd. *et al*, 1931].

TABLE V
Showing numbers and duration of pollen-collecting trip of *Andrena ilderda* Cam. during favourable weather

No. of observa- tion	Date	Time		Duration of						Remarks.
		Left burrow	Returned to burrow	First visit		Second visit		Third visit		
				hrs.	mts.	hrs.	mts.	hrs.	mts.	
I	29-11-1930	10-13 A.M.	1-23 P.M.	3	10	Observation discontinued after 3-35 P.M.
		1-31 P.M.	3-35 P.M.	2	4	
II	1-12-1930	10-36 A.M.	12-13 P.M.	1	37	Returned at 12-13 being hotly pursued by <i>Philanthus depredator</i> . Took shelter under a lump of earth and on removal of clod ran into the burrow. Did not return up to 5-30 P.M. when observation was discontinued.
		12-19 P.M.	1-45 P.M.	1	26	
		1-48 P.M.	
		/	
III	2-12-1930	10-9 A.M.	1-14 P.M.	3	5	Did not come out upto 5-30 P.M. when observation was discontinued. Caught in a tube placed at the entrance next day.
		1-24 P.M.	3-46 P.M.	2	22	
IV	8-12-1930	3-50 P.M.	5-11 P.M.	1	21	Observation discontinued after 4-49 P.M.
		2-10 P.M.	4-49 P.M.	2	39	
V	20-12-1930	10-19 A.M.	2-46 P.M.	4	27	Observation discontinued after 5-30 P.M. as the insect did not return.
		2-55 P.M.	4-48 P.M.	1	53	
		4-50 P.M.	5-0 P.M.	0	10	
VI	22-12-1930	3-6 P.M.	4-47 P.M.	1	41	Observation discontinued after 4-47 P.M.

Like *Apis florea* Fab. *Andrena ilderda* Cam. also works in a purposeful manner and table VI gives the number of flowers visited in one minute.

TABLE VI

Showing number of flowers visited per minute by Andrena ilderda Cam.

Date	Time of observation		Duration of observation (minutes)	Total number of flowers visited	Number of flowers visited in one minute
	From P. M.	To P. M.			
26-11-30	1.28	1.30	2	13	6.5
26-11-30	2.46	2.49	3	18	6
28-11-30	1.34	1.36	2	15	7.5
4-12-30	12.15	12.19	4	33	8.2
16-12-30	1.30	1.33	3	26	8.6
19-12-30	2.40	2.42	2	16	8
27-11-31	3.6	3.11	5	39	7.8
30-11-31	1.15	1.17	2	15	7.5
5-12-31	1.15	1.18	3	22	7.3
14-12-31	12.45	12.49	4	32	8

Thus during favourable weather *Andrena ilderda* Cam., on an average, visits 7.5 flowers per minute.

A. ilderda Cam. is the most important insect for *toria* pollination, but being wild it is not possible to do anything with it in a practical way to ensure its presence, or augment its numbers, in *toria* fields.

At Lyallpur the appearance of *Halictus* sp. in *toria* synchronizes with its (*toria*) flowering: it remains active in this crop until *sarson* flowers appear. Thus before the appearance of *Andrena ilderda* Cam. it plays the most important part in the pollination of *toria*.

In its nesting habits it resembles *Andrena ilderda* Cam. It lives in small colonies, each member occupying a cell which is joined to the main tunnel by a side branch.

Table VII gives the number of flowers visited by *Halictus* in one minute.

TABLE VII

Showing number of flowers visited per minute by *Halictus* sp.

Date	Time of observation		Duration of observation (minutes)	Total number of flowers visited	Number of flowers visited in one minute
	From P. M.	To P. M.			
17-1-30	12.0	12.8	8	20	2.5
26-1-30	2.58	3.3	5	18	3.6
19-2-30	2.10	2.14	4	14	3.5
22-2-30	1.25	1.28	3	17	5.6
16-11-31	1.30	1.34	4	13	3.2
20-11-31	1.46	1.49	3	7	2.3
30-11-31	2.11	2.17	6	23	3.8
11-12-31	2.1	2.3	2	8	4

Thus during favourable weather *Halictus* sp., on an average, visits 3.5 flowers per minute.

Fam. COLLETIDÆ.—*Colletes reticulata* (Cam.), *C. nursei* Cam., and *Colletes* sp. are the only representatives of these primitive bees which were taken from *sarson* flowers. Only six specimens were collected and this fact alone marks them out to be useless as pollinating agents.

Fam. XYLOCOPIDÆ.—*Xylocopa* (*Nyctomelita*) *nasalis nasalis* Westw. is the only representative of this family secured from *toria* and *sarson* flowers. It is a powerful flier producing a loud buzzing noise during flight. It is never abundant—only eight specimens were collected—and this fact alone rules it out from the list of useful pollinators.

Fam. CERATINIDÆ.—*Ceratina sexmaculata* Sm. is a casual visitor to *toria* and *sarson* flowers. It is a slow and lazy worker and its method of obtaining nectar is such that only the front of its head comes in contact with the anthers. It has no value as a pollinating agent.

Fam. NOMADIDÆ.—Of the three species collected *Nomada* 2 spp. are comparatively more abundant as is seen from the following table :—

TABLE VIII

Showing names and numbers of *Nomadidæ* collected from *toria* and *sarson* during 1930 and 1931

Name	1930		1931	
	<i>toria</i> (November 19-25 December 1-7)	<i>sarson</i> (January 13-19 February 13-19)	<i>toria</i> (November 11-17 December 7-13)	<i>sarson</i> (January 24-30 February 13-19)
<i>Nomada</i> 2 spp.	65	38	163	23
<i>Crocisa ramosa</i> Lep. ?	1	..

The general appearance of *Nomada* 2 spp. is suggestive of *Polistes hebræus* (Fab.)

During November and December, they were often seen at 8 A.M. suspended from leaves of *toria* by their mandibles. They are poor fliers and always fly low to the ground. They live as parasites in the nests of *Andrena* spp. and *Halictus* spp. for their larvæ are fed 'upon the provisions originally destined for the progeny of the host species' [Imms, 1934]. They are, therefore, undesirable visitors to *toria* and *sarson* fields.

Fam. ANTHOPHORIDÆ.—*Anthophora vedetta* Nurse has the longest proboscis among the bees discussed in this paper. It prefers to feed in flowers where the nectaries are deep seated, e.g., 'taramira' (*Eruca sativa*). It is, therefore, very rarely that one collects it from *sarson*.

Fam. BEMBECIDÆ.—*Bembex trepanda* Dahlb. is the only representative of this family taken from *toria* and *sarson* flowers.

It nests in the ground and shows parental care for its offspring. It preys upon Syrphidæ, Muscidæ and Calliphoridæ which it supplies daily to its larvæ. It is therefore an undesirable visitor to *toria* and *sarson* flowers.

Fam. LARIIDÆ.—*Liris hæmorrhoidalis* (Fab.) is the only representative of this family secured from *toria* flowers. It preys upon *Andrena ilderda* Cam. and other bees and is therefore an undesirable visitor to *toria* flowers.

Fam. PHILANTHIDÆ.—*Philanthus depredator* Smith is the only representative of this family secured from *toria* flowers.* Table IX gives the numbers of this insect collected from *toria* flowers during the specified weeks.

* It is in hibernation during the flowering period of *sarson*.

TABLE IX

Showing numbers of *Philanthus depredator Smith* collected from *toria* during 1930 and 1931

Name	1930	1931
	<i>toria</i> (November 19-25 December 1-7)	<i>toria</i> (November 11-17 December 7-13)
<i>Philanthus depredator Smith</i>	90	118

P. depredator Smith is a handsome and graceful insect. It constructs sinuous burrows in the soil. It closes the burrow entrance before going out to 'hunt'. This is done by shovelling back the soil particles with its legs 'in the manner of a dog digging'. On its return the prey is at first deposited on the ground near to the entrance hole which is then opened and the prey dragged into the burrow.

P. depredator Smith alternates its work of destruction with that of feeding on nectar. It is, however, a very slow and unsteady worker as is seen from Table X.

TABLE X

Showing number of flowers visited per minute by *Philanthus depredator Smith*

Date	Time of observation		Duration of observation (minutes)	Total number of flowers visited	Number of flowers visited in one minute
	From	To			
20-11-30	P.M. 2.1	P.M. 2.3	2	8	4
23-11-30	2.16	2.21	5	20	4
29-11-30	2.16	2.21	5	18	3.6
1-12-30	1.0	1.7	7	29	4.1
21-11-31	12.0	12.2	2	9	4.5
27-11-31	1.4	1.7	3	14	4.6
30-11-31	1.15	1.20	5	23	4.6

Thus, on an average, this insect visits about four flowers per minute.

Its main object in visiting *toria* flowers, however, is to prey upon *Andrena ilerda* Cam., and *Apis florea* (Fab.), etc. for provisioning its nest.

P. depredator Smith attacks a bee when it (bee) is busy collecting pollen and nectar, and on average takes 30 seconds (average of 12 observations) to paralyse it. Afterwards, it is 'carted' to the burrow in the usual Sphegid manner. Thus *P. depredator* Smith is the most unwelcome visitor to *toria* flowers.

Fam. VESPIDÆ.—Impregnated females of *Polistes hebraeus* (Fab.) and *Vespa orientalis* * Linn. visit *toria* flowers during November and 1st week of December.

They are predaceous upon Apidæ and Andrenidæ: they feed upon their honey-stomachs which they obtain by biting off the thorax. They are thus undesirable visitors to *toria* flowers.

Fam. EUMENIDÆ.—Of the three species collected *Odynerus* sp. was comparatively more abundant as is seen from the following table:—

TABLE XI

Showing names and numbers of Eumenidæ collected from *toria* and sarson during 1930 and 1931

Name	1930		1931	
	<i>toria</i> (November 19-25 December 1-7)	<i>sarson</i> (January 13-19 February 13-19)	<i>toria</i> (November 11-17 December 7-13)	<i>sarson</i> (January 24-30 February 13-19)
<i>Odynerus</i> sp.	16	..	26	..
<i>Eumenes dimidiatipennis</i> Sauss.	2	..
<i>Eumenes</i> sp.	1

Odynerus sp. is a slenderly built insect. It visits *toria* flowers to feed on nectar and pollen. Because of its smooth body it is useless as a pollinating agent.

The female possesses a formidable sting. It is used to sting Lepidopterous larvæ into a state of torpor for storing them in a specially constructed earthen cell as food for its progeny.

Fam. SCOLIDÆ.—*Campsomeris thoracicus* (Fab.), *C. thoracicus* var. *aureicollis* Lep. and *Scolia* sp. are the only representatives of this family secured from *toria* flowers. They are rapid fliers and are difficult to catch. During work they show a certain degree of persistency and fly rather than crawl.

* It is interesting to record here that on 22-11-31 at 3-14 P.M. a King crow (*Dicrurus macrocerus macrocerus* Vieill.) was observed to snap at and swallow three *Vespa orientalis* Linn. within five minutes.

from flower to flower. They obtain nectar like honey bees. But they are never abundant, therefore, their value as pollinating agents is limited.

Fam. CHRYSIDIDÆ.—Three specimens of *Chrysis indica* Macs. were collected from '*toria*' flowers. It has a brilliant metallic green colour and when disturbed it rolls itself quickly into a ball. It is a parasite of *Bembex*, *Odynerus* and *Eumenes*.

Fam. FORMICIDÆ.—Only two representatives, namely *Cataglyphis bicolor* subsp. *Setipes* (Forel), and *Messor barbarus* (Linn.) var. *instabilis* Sm. of this family were found visiting *toria* and *sarson* flowers to feed on nectar.

During feeding *C. bicolor* subsp. *Setipes* (Forel) introduces its head from between the floral 'claws' to reach the nectaries, the body sprawling over the adjacent flowers. When frightened it drops to the ground. Its body is smooth and unfit for the carriage of pollen.

Winged forms of *Messor barbarus* Linn. var. *instabilis* Sm. visit *toria* and *sarson* flowers but are never abundant. Their body being covered with hairs is very well suited for the carriage of pollen. Therefore, they cannot be entirely useless as pollinating agents.

Order DIPTERA

Fam. SYRPHIDÆ.—This family was strongly represented as is seen from the following table:—

TABLE XII

Showing names and numbers of *Syrphidæ* collected from *toria* and *sarson* during 1930 and 1931

Name	1930		1931	
	<i>toria</i> (November 19-25 December 1-7)	<i>sarson</i> (January 13-19 February 13-19)	<i>toria</i> (November 11-17 December 7-13)	<i>sarson</i> (January 24-30 February 13-19)
<i>Eristalis tenax</i> (Linn.)	28	66	19	74
<i>E. quinquelineatus</i> (Fab.)	10	13	18	8
<i>E. ceneus</i> Scop.	8	7	11	2
<i>E. tæniops</i> Wied.	4	4	..	3
<i>Sphaerophoria indiana</i> Big.	3	4	4	1
<i>Syrphus balteatus</i> (de Geer)	6	1
<i>Lasipticus albomaculatus</i> Macq.	1	1
<i>Ichiodon scutellaris</i> (Fab.)	3

Eristalis tenax (Linn.) is a heavy bodied insect which bears a close resemblance to *Apis indica* Fab. During flight it produces a loud hum. It is a graceful insect to watch on the wing, but its movements on the flowers are awkward and clumsy. It is a spasmodic worker because 'hovering' over a flower and sitting and cleaning itself are as important to it as feeding.

Its *modus operandi* on a flower is identical with that of *Apis florea* Fab. described before. Its progress on flowers is very satisfactory as is seen from Table XIII.

TABLE XIII

Showing number of flowers visited per minute by *Eristalis tenax* Linn.

Date	Time of observation		Duration of observation (minutes)	Total number of flowers visited	Number of flowers visited per minute
	From	To			
18-1-30	P.M. 3.5	P.M. 3.12	7	47	6.7
25-1-30	2.7	2.10	3	18	6
7-2-30	1.38	1.49	11	97	8.8
19-11-30	12.10	12.18	8	49	6.1
8-12-30	A.M. 11.4	A.M. 11.8	4	39	9.7
13-12-30	P.M. 1.58	P.M. 2.1	3	26	8.6
11-2-30	1.15	1.18	3	15	5
11-11-31	A.M. 10.9	A.M. 10.15	6	35	5.8
24-11-31	11.11	11.20	9	60	6.6
7-12-31	P.M. 1.5	P.M. 1.10	5	34	6.8

Thus, on an average, it visits seven flowers per minute.

It is found in the fields from early morning to late afternoon and is met with in all types of weather. It is an important pollinator.

The other species of *Eristalis* though much less abundant are equally efficient as pollinating agents. *Sphærophoria indiana* Big. is a small sized insect which hovers 'poised motionless' in the air over a flower before alighting. It is a spasmodic worker.

Lasioticus albomaculatus Macq., *Ischiodon scutellaris* Fab., and *Syrphus balteatus* de G. are of little significance as pollinating agents. The larvæ of the last named, however, destroy the Mustard Aphid.

Fam. AGROMYZIDÆ.—*Phytomyza atricornis* Meig., a representative of this family, is a minute insect which is quite abundant towards the end of *sarson* season. When disturbed it shoots up in the air and looks like a dust particle.

It is found on the wing in the mornings, evenings and on cloudy days, but has never been observed visiting *sarson* flowers. When it is sunny and warm it takes shelter under cover of the leaves lying on the ground or other suitable shady places.

Its larvæ damage *sarson* leaves by making zigzag whitish galleries. When infestation is severe every leaf may be affected. The attacked leaf becomes distorted, turns yellow and ultimately falls down.

Fam. MUSCIDÆ.—Six species, namely, *Musca domestica nebulo vicina* Macq., *M. vitripennis* Meig., *M. domestica nebulo* Fab., together with three undetermined species of the genus *Musca* are the only representatives of this family secured from the flowers of the two crops.

Musca domestica nebulo vicina Macq. is an erratic and an unstable worker, and is not a persistent seeker of pollen and nectar. It also licks up stigmatic secretions. When feeding on nectar its body usually lies perpendicularly between the stalks of the stamens and the style of the stigma, but when licking up stigmatic secretions or feeding on pollen its body usually rests on petals. After every 'feed' it may settle on the body of the observer, on bags, bamboos, plants, or on the ground, or may even engage in a playful 'combat' with another 'fellow'. But it is found (along with other Muscids) in the fields from morning till late in the afternoon and is active both during fine and inclement weather. Therefore, it is safe to infer that Muscids in general and *Musca domestica nebulo vicina* Macq. in particular may play a minor part in pollination.

Fam. CALLIPHORIDÆ.—Of the six species of this family collected *Trichometallea pollinosa* Tns. was consistently more abundant as is seen from Table XIV.

TABLE XIV

Showing names and numbers of Calliphoridae collected from toria and sarson during 1930 and 1931

Name	1930		1931	
	toria (November 19-25 December 1-7)	sarson (January 13-19 February 13-19)	toria (November 11-17 December 7-13)	sarson (January 24-30 February 13-19)
<i>Trichometallea pollinosa</i> Tns.	24	14	19	29
<i>Rhinia discolor</i> Fab.	4	3	29	6
<i>Lucilia sericata</i> Meig.	2	..	5	1
<i>Calliphora erythrocephala</i> Meig.	..	5	..	1
<i>Sarcophaga</i> sp.	5	..
<i>Chrysomya megacephala</i> Fab.	1

Trichometallea pollinosa Tns. feeds on nectar and pollen as well as licks up stigmatic secretions. Its habits and mode of feeding resemble those of *Musca domestica nebulosa* vicina Macq. When working seriously its rate of progress is fairly satisfactory (Table XV).

TABLE XV

Showing number of flowers visited per minute by *Trichometallea pollinosa* Tns.

Date	Time of observation		Duration of observation (minutes)	Total number of flowers visited	Number of flowers visited per minute
	From	To			
	A.M.	A.M.			
18-1-30	10.12	10.16	4	18	4.5
21-1-30	11.39	11.43	4	11	2.75
	P.M.	P.M.			
25-1-30	3.0	3.2	2	8	4
	A.M.	A.M.			
25-11-30	10.48	10.53	5	21	4.2
26-11-30	10.0	10.3	3	9	3
	P.M.	P.M.			
1-12-30	12.30	12.32	2	11	5.5
6-2-31	2.30	2.33	3	7	2.3
17-2-31	1.7	1.9	2	7	3.5
20-11-31	1.18	1.22	4	15	3.75
15-12-31	1.17	1.19	2	12	6

Thus it visits, on an average, 3.9 flowers per minute.

It is a fairly useful insect as a pollinating agent.

Rhinia discolor Fab. is about the size of a house-fly, but is darker in colour with whitish abdomen. It hovers over a flower like a Syrphid pinned to the spot: it may, however, dart to one side, disappear momentarily and then reappear about the same spot. During flight it produces a characteristic buzzing sound.

Its habits and mode of feeding resemble those of *T. pollinosa* Tns. Because of its numbers its value as a pollinating agent cannot be questioned.

L. sericata Meig., *C. erythrocephala* Meig., and *C. megacephala* Fab. are too few in *toria* and *sarson* flowers to be of any use as pollinating agents.

Fam. SEPSIDÆ.—Of the two species collected, *Sepsis* sp. was commoner as is seen from Table XVI.

TABLE XVI

Showing names and numbers of *Sepsidæ* collected from *toria* and *sarson* flowers during 1930 and 1931

Name	1930		1931	
	<i>toria</i> (November 19-25 December 1-7)	<i>sarson</i> (January 13-19 December 7-13)	<i>toria</i> (November 11-17 December 7-13)	<i>sarson</i> (January 24-30 February 13-19)
<i>Sepsis</i> sp.	..	98	3	66
<i>S. ephippium</i> Bezzi	..	7	..	2

Sepsis sp. is a slenderly built insect. After every 'feed' it may clean its smooth body for as long as nine minutes before visiting another flower. It is a sluggish and an erratic worker but because of its abundance its value as a pollinating agent cannot be doubted.

Fam. CONOPIDÆ.—*Conops erythrocephala* Fab. is the only representative of this family caught in November from *toria* flowers. Its larvæ are endoparasites of *Andrena* and *Vespa*. It is, therefore, an unwelcome visitor to *toria* flowers.

Fam. ORTALIDÆ.—*Chrysomya demandata* Fab. is the only representative of this family secured from *toria* and *sarson* flowers. Because of its very small numbers, it is useless as a pollinating agent.

Fam. CORDYLURIDÆ.—Only two specimens of *Scatophaga* sp. were secured from *sarson* in 1930. It is useless as a pollinating agent.

Fam. TRYPETIDÆ.—*Dacus zonatus* Saund. is the representative of this family that was secured only once from *toria* flowers. It is useless as a pollinating agent.

Fam. EMPIDÆ.—*Hilara* sp. is the only representative of this family of predaceous flies collected from *sarson* which, because of its very rare occurrence, is useless as a pollinating agent.

Fam. MILICHIIDÆ.—*Desmometopa M-nigrum* Zett. is the only representative of this family which was collected only once. It is useless for pollination.

Order LEPIDOPTERA

Names and numbers of the Butterflies collected from *toria* are given below: they were never found visiting *sarson* flowers.

TABLE XVII

Showing names and numbers of Butterflies collected from toria during 1930 and 1931

Name	1930	1931
	<i>toria</i> (November 19-25 December 1-7)	<i>sarson</i> (November 11-17 December 7-13)
DANAIDÆ :		
<i>Danaus chrysippus</i> (Linn.)	6	11
NYMPHALIDÆ :		
1. <i>Precis orithya</i> Linn.	..	1
2. <i>Vanessa cardui</i> (Linn.)	1	2
3. <i>Hypolimnas misippus</i> Linn.	..	1
PIERIDÆ :		
1. <i>Catopsilia crocale</i> Cram.	5	7
2. <i>Anaphæis mesentina</i> Cram. = <i>aurora</i> Fab.	5	4
3. <i>Pieris brassicæ</i> Linn.	1	2
LYCÆNIDÆ :		
<i>Polyommatus</i> sp.	1	..
HESPERIIDÆ :		
<i>Badamia exclamationis</i> Fab.	1	1

The Butterflies mentioned in Table XVII above visit flowers for feeding on nectar.

To obtain nectar a Butterfly suspends itself from petals and simultaneously probes the nectaries with its extended proboscis. To reach the next flowers it may either 'flutter' across the entire bunch of flowers or may fly to it. On an average a Butterfly may visit one flower in one minute.

Because of their hairy bodies and presence both during fine and inclement weather the value of Butterflies as pollinating agents cannot be doubted.

Fam. PYRALIDÆ.—*Noctuelia floralis* Hubn. is the only representative of this family secured from *toria* and *sarson* flowers. It is nocturnal and whether it visits these flowers in sufficiently large numbers at night to play any part in their pollination yet remains to be ascertained.

Fam. ARCTIIDÆ.—*Utetheisa pulchella* Linn. was about as common as *D. chrysippus* Linn. During feeding it crawls over the flowers rather sluggishly; as such its value as a pollinating agent cannot be doubted.

Fam. NOCTUIDÆ.—*Earias insulana* (Bosid.) and *Laphygma exigua* Hub. are the only representatives of this family secured from *toria* and *sarson* flowers respectively. They were evidently accidental visitors.

A single caterpillar of *Plusia chalcytes* Fab. was found damaging *toria* flowers in December, 1930.

Fam. LYMANTRIIDÆ.—*Euproctis* sp. was secured from *toria* flowers only once.

Order COLEOPTERA

Fam. CARABIDÆ.—One specimen of *Pterostichus leus* Andr.—a representative of this family of nocturnal and predaceous beetles—was secured in November.

Fam. NITIDULIDÆ.—*Hoptoncus lutelus* Chevr. is the only member of this family of which two specimens were collected from *toria* and *sarson* flowers.

Fam. COCCINELLIDÆ.—Four species namely, *Chilomenes sexmaculata* Fab., *Coccinella septempunctata* (Linn.), *C. undecimpunctata* Linn. and *Adonia variegata* subsp. *doubledayi* Muls. of this family were secured from *toria* and *sarson* flowers. They usually abound in *toria*.

Larvae and adults of these predaceous beetles move about actively among *toria* and *sarson* flowers in search of their prey, e.g., Aphididae. The larvae are exclusively carnivorous, but the adults alternate their insect food with nectar. Therefore the greatest good they do is to destroy the injurious Mustard Aphis (*Siphocoryne indobrassicae* Das). Also, because of their body structure and assiduity with which they search out their prey, their value in the transfer of certain amount of pollen cannot be doubted.

Fam. DERMESTIDÆ.—*Attagenus bifasciatus* Oe. is the only representative of this family which has been secured from *toria* flowers.

It feeds on nectar and as many as five of them may be found in the same flower. During feeding its body lies parallel to the style with its head directed towards the nectaries. It may remain in this position for as long as 12 minutes.

It visits *toria* flowers in January when the crop is ready to be harvested. This fact coupled with its sedentary habits marks it out to be useless as a pollinating agent.

Fam. MELYDRIDÆ (*Malachiidæ*).—A single specimen of *Laius malleifer* Champ.—a representative of this family—was collected in November.

Fam. TENEBRIONIDÆ.—A single specimen of *Opatroides vicinis* Fairm.—a representative of this family—was collected in January.

Fam. CHRYSOMELIDÆ.—About six specimens of the dreaded *Aulacophora foveicollis* Luccap. were collected from November to February. This insect

winters over as an adult and therefore the specimens which were collected had evidently come out to 'enjoy' the sunshine.

Fam. CURCULIONIDÆ.—Two specimens of *Mylocerus maculosus* Desb. were collected in December. Like *Aulacophora* this insect also winters over as an adult.

Order NEUROPTERA

Fam. CHRYSOPIDÆ.—*Chrysopa* sp. is the only representative of this family which has been collected from the two crops. Though useless as a pollinating agent, its larva does good work in destroying the Mustard Aphis, each larva killing 25-28 Aphids per day.

Order ORTHOPTERA

Fam. MANTIDÆ.—*Creobroter gemmatus* Stoll. is the only representative of this family secured from flowers. It preys upon insect visitors to *toria* and *sarson* flowers and is, therefore, an undesirable creature.

Order ODONATA

Fam. LIBELLULIDÆ.—*Pantala flavescens* Linn.—a representative of this family—was occasionally seen settled on *toria* and *sarson* plants. Its visits were purely for the purpose of preying upon insects making up the fauna of the two crops but what precisely they preyed upon could not be ascertained.

Order THYSANOPTERA

Fam. AEOLOTHRIPIDÆ.—(?) *Aeolothrips* sp. is the only representative of this family collected from *toria* and *sarson* flowers. It feeds both on nectar and sap of flowers: it obtains the latter substance by lacerating the flower tissue with its rasping-sucking mouth-parts.

Order RHYNCHOTA

Fam. PENTATOMIDÆ.—The Painted Bug, *Bagrada picta* (Fab.), is a serious pest of *Brassicae*. It is fairly common in *toria* in November where it is seen settled on flower-buds sucking sap with its stylets. Seriously attacked flower-buds open badly and imperfectly.

Dolycoris indicus Stal. and *Nezara viridula* (Linn.) were less abundant and of absolutely no value as pollinating agents.

Fam. COREIDÆ.—One specimen of *Liorhyssus hyalinus* Fab. collected in February from *sarson* was evidently an accidental visitor and as such useless as a pollinating agent.

Fam. LYGAEIDÆ.—One specimen each of *Spilostethus pandurus* (Scop.), *Graptostethus servus* (Fab.) and *Oxycarenus laetus* Kirby was collected in December, January and February respectively. Because of their small numbers Lygaeids are of no importance in pollination.

Fam. PYRRHOCORIDÆ.—The Red Cotton Bug, *Dysdercus cingulatus* (Fab.), is very rarely met with in *toria* and *sarson* and is, therefore, useless as a pollinating agent.

Fam. MEMBRACIDÆ.—The common Tree Hopper, (?) *Leptocentrus* sp. which abounds on 'Sirin' (*Albizzia lebbek*) was collected only once in November.

Fam. FULGORIDÆ.—The sugarcane Leaf Hopper, *Pyrilla perpusilla* (Walk.), is a pest of sugarcane and wheat: the two specimens collected in November were evidently accidental visitors.

Fam. APHIDIDÆ.—The common Mustard Aphis, *Siphocoryne indobrassicae* Das. (= *pseudobrassicae* Davis) abounds on *toria* and *sarson*. It congregates in enormous numbers on the stalk of the inflorescence, flower-buds and flowers from which it sucks vital fluids with its stylets. The attacked stalk gnarls badly and the flowers open poorly, while the pods formed from such flowers are misshapen and usually do not develop any seed. It is, therefore, a serious pest whose absence rather than presence is to be desired.

RELATIVE SIGNIFICANCE OF THE IMPORTANT POLLINATORS

A method of study.—It will be observed from the foregoing pages that the insect visitors of *toria* and *sarson* flowers constitute a complex phenomenon of organic activity. Some are sap-feeders, e.g., *Bagrada picta* (Fab.) *Siphocoryne indobrassicae* Das. (= *pseudobrassicae* Davis.), etc., some are predaceous upon other insects, e.g., *Creobroter gemmetus* Stoll., *Liris haemorrhoidalis* (Fab.), *Philanthus depredator* Smith, etc., a few are parasites of other insects, e.g. *Nomada* sp., a few are accidental visitors to these crops, e.g. *Dysdercus cingulatus* (Fab.), *Pyrilla perpusilla* Walk., etc., whilst a vast majority (particularly Hymenoptera and Diptera) visit these flowers for pollen and nectar.

To study the relative significance of the insect visitors to *toria* and *sarson* flowers as pollinating agents the following method was adopted:—

Two healthy shoots of the same age were selected on an easily accessible plant and their opened and about to open flowers were nipped off with scissors. One of these shoots was used for cross-pollination by insects and the other as control. The flower-buds were carefully examined for Thrips and other small insects. Each branch was then enclosed in a muslin bag (30 in. by 15 in.) big enough to provide ample space for its growth. The top of the bag was fastened to an inverted L shaped bamboo stick hammered in the ground. The bags thus secured are able to stand a casual storm which may occur during the growing season of *toria* or *sarson*. Six insects were introduced daily between 12 noon to 2 P.M. into each bag. These insects were captured in glass tubes and immediately put into the bags from the upper end. At the close of each experiment all the unopened flowers enclosed in a bag were nipped off and the bag tied up in the usual manner so as to allow the seeds to mature. In due course the plants were harvested and the seeds formed in each pod were counted. The results are presented in Table XVIII. (Only a typical example is given in each case).

It will be observed from Table XVIII that the largest number of normal seeds per pod was found in bags with *Andrena ilderda* Cam. and *Apis florea* Fab., while the lowest number of seeds was formed in bags with *Sepsis* sp. and *Trichometallea pollinosa* Tns.

It will also be observed from Table XVIII that the average number of normal seeds formed per pod in the case of the experimental plants was



TABLE XVIII

Showing relative significance of the insect pollinators a : bag with insects. b : Control. c : free flowering

	<i>Andrena ilenia</i> Cam.			<i>Apis florea</i> F.			<i>Halictus</i> sp.			<i>Philaethus degeerdator</i> Smith			<i>Eristalis</i> sp.			<i>Sepsis</i> sp.			<i>T. polinosa</i> Tns.		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
Total flowers	118	111	73	215	303	50	68	66	53	108	170	85	176	124	30	26	21	14	220	166	55
Pods formed	115	22	73	202	45	50	58	34	51	40	16	78	139	51	28	5	4	13	102	53	54
Per cent pod setting	97.46	19.82	100.0	93.98	14.85	100.0	85.99	51.51	96.28	37.0	9.4	91.8	78.98	41.13	93.33	19.2	19.0	92.8	46.4	31.9	93.2
Average No. of normal seed per pod.	7.53	1.42	18.51	6.95	1.54	18.02	3.68	1.88	20.02	1.1	0.8	12.2	4.38	2.07	28.32	1.2	0.8	10.7	1.6	1.7	17.2

TABLE XIX
Showing the total number of insects collected during November-March, 1930 and 1931.

	1930			1931		
	toria		salson	toria		salson
	No.	Per cent	No.	Per cent	No.	Per cent
HYMENOPTERA—						
<i>Andrena ilderda</i> . . .	586	43.73	76	5.35	1420	39.78
<i>Apis florea</i> . . .	221	16.49	690	48.62	684	19.16
<i>Philanthus depredator</i> . . .	90	6.71	118	3.30
<i>Halictus</i> sp. . .	52	3.88	89	6.27	367	10.28
<i>Nomada</i> spp. . .	65	4.85	38	2.67	163	4.56
Other Hymenoptera . . .	98	7.31	60	4.22	183	5.12
DIPTERA—						
<i>Eristalis</i> spp. . .	50	3.73	90	6.34	48	1.34
<i>Trichometathea pollinosa</i> . . .	64	4.77	74	5.21	62	1.73
<i>Sepsis</i> sp.	211	14.86	16	.44
<i>Musca</i> sp.	5	.35	35	.98
<i>Rhinia discolor</i> . . .	4	.29	3	.21	135	3.78
Other Diptera . . .	49	3.65	58	4.08	146	4.09
LEPIDOPTERA . . .	10	.74	16	1.12	66	1.84
COLEOPTERA . . .	23	1.71	4	.28	60	1.68
RYNCHOTA . . .	31	2.31	48	1.34
NEUROPTERA . . .	1	.07	15	.42
THYSANOPTERA	8	.56
ORTHOPTERA	3	.08
Total . . .	1344	..	1422	..	3569	..
GRAND TOTAL . . .	7491

TABLE
Showing names and numbers

<i>Toria</i> flowers (14-12-1931)	8-40—9 hrs.	9—10 hrs.	10—11 hrs.	11—12 hrs.
Inflorescence with 18 flowers	<i>Trichometallea pollinosa</i> . 1	<i>Rhinia discolor</i> . 1	<i>Andrena ilerda</i> . 2	<i>Andrena ilerda</i> . 5 <i>Apis florea</i> . 2 <i>Musca</i> sp. . 1 <i>Anthophora vedetta</i> . 1
A marked flower of the above inflorescence	<i>Trichometallea pollinosa</i> . 1	...	<i>Andrena ilerda</i> 2 <i>Anthophora vedetta</i> . 2	...
Single flower, others clipped off.	<i>Andrena ilerda</i> . 8	<i>Andrena ilerda</i> . 5 <i>Musca domestica nebulo</i> . 1 <i>Eristalis</i> sp. . 1 <i>Trichometallea pollinosa</i> . 1
<i>Sarson</i> flowers (19-2-1931)				
Inflorescence with 20 flowers	<i>Eristalis</i> sp. . 2 <i>Sepsis</i> sp. . 1	<i>Musca</i> sp. . 5 <i>Sepsis</i> sp. . 3 <i>Halictus</i> sp. . 1	<i>Musca</i> sp. . 5 <i>Sepsis</i> sp. . 1	<i>Apis florea</i> . 8 <i>Sepsis</i> sp. . 4 <i>Andrena ilerda</i> . 1
A marked flower of the above inflorescence.	...	<i>Sepsis</i> sp. . 3	<i>Halictus</i> sp. . 1	<i>Apis florea</i> . 2
Single flower, others clipped off.	<i>Musca</i> sp. . 2 <i>Rhinia discolor</i> . 1	<i>Eristalis</i> sp. . 1	...	<i>Apis florea</i> . 2 <i>Sepsis</i> sp. . 1

XX

of insects visiting between

12—13 hrs.	13—14 hrs.	14—15 hrs.	15—16 hrs.	16—17 hrs.
<i>Andrena ilderda</i> . 13	<i>Andrena ilderda</i> . 28	<i>Andrena ilderda</i> . 25	<i>Andrena ilderda</i> . 8	<i>Sphaerophoria</i> <i>indiana</i> . . 1
<i>Apis florea</i> . . 3	<i>Halictus</i> sp. . 1	<i>Chrysomya</i> <i>demandata</i> . 1	<i>Apis florea</i> . . 2	<i>Anthophora</i> <i>vedetta</i> . . 1
<i>Eristalis</i> sp. . 2	<i>Apis florea</i> . 1	<i>Musca</i> sp. . 1	<i>Musca</i> sp. . 2	
<i>Musca</i> sp. . . 1	<i>Musca domestica</i> <i>nebulo</i> . . 1		<i>Eristalis</i> sp. . 2	
	<i>Rhynchota</i> . 1		<i>Philanthus depre-</i> <i>dator</i> . . 1	
....	<i>Andrena ilderda</i> . 4	<i>Andrena ilderda</i> . 6	<i>Philanthus depre-</i> <i>dator</i> . . 1	...
	<i>Apis florea</i> . 1			
<i>Andrena ilderda</i> . 3	<i>Andrena ilderda</i> . 10	<i>Andrena ilderda</i> . 4	<i>Andrena ilderda</i> . 4	<i>Rhinia discolor</i> . 1
	<i>Apis florea</i> . 3	<i>Apis florea</i> . 2	<i>Apis florea</i> . 1	
			<i>Eristalis</i> sp. . 1	
<i>Apis florea</i> . . 17	<i>Apis florea</i> . 21	<i>Apis florea</i> . 21	<i>A. florea</i> . . 9	<i>A. florea</i> . . 2
<i>Sepsis</i> sp. . 7	<i>Halictus</i> sp. . 4	<i>Sepsis</i> sp. . . 3	<i>Sepsis</i> sp. . . 6	<i>Sepsis</i> sp. . . 2
			<i>Musca</i> sp. . . 2	
<i>Musca</i> sp. . . 2	<i>Trichometalla</i> <i>polliniosa</i> . 2	<i>A. ilderda</i> . . 2	<i>A. ilderda</i> . . 1	
<i>Andrena ilderda</i> . 2	<i>Sepsis</i> sp. . . 2	<i>Halictus</i> sp. . 1	<i>Halictus</i> sp. . 1	
<i>Halictus</i> sp. . 2	<i>Andrena ilderda</i> . 1	<i>Musca</i> sp. . 1		
	<i>Eristalis</i> sp. . 1	<i>Eristalis</i> sp. . 1		
<i>Apis florea</i> . . 1	<i>Apis florea</i> . 3	<i>A. florea</i> . . 3	...	<i>A. florea</i> . . 2
<i>Andrena ilderda</i> . 1	<i>Andrena ilderda</i> . 1	<i>Halictus</i> sp. . 1		
<i>A. florea</i> . . 4	<i>A. florea</i> . . 5	<i>A. florea</i> . . 3	<i>A. florea</i> . . 3	<i>Musca</i> sp. . 1
<i>A. ilderda</i> . . 1	<i>A. ilderda</i> . . 2		<i>A. ilderda</i> . . 1	
<i>Sepsis</i> sp. . . 1	<i>Sepsis</i> sp. . . 2			

considerably below that of the free-flowering plants. This was to be expected. When introduced (or rather 'imprisoned') in a bag an insect flew about excitedly so as to discover an exit for escape. By the time it settled down very little pollen was left on its body to ensure complete pollination.

Table XVIII (Control bags) also shows that *toria* and *sarson* plants are not entirely self-sterile for a certain amount of self pollination does take place in these two plants. This corroborates the observations of Ali Mohd. *et al.* [1931].

Table XIX gives the total number of insects collected during November-March, 1930 and 1931, as well as the names of those insects which predominated amongst the insect visitors, the rest being lumped together under their respective orders. Numbers and percentages, however, are given in all cases.

A study of Table XIX justifies the following conclusions :—

1. *Andrena ilerda* Cam. constitutes 40-44 per cent of the insect visitors to *toria* flowers. In *sarson* it makes up 3.5 per cent only of the insect visitors.
2. *Apis florea* Fab. constitutes 49-52 per cent of the visitors to *sarson*, but only 16-17 per cent to *toria* flowers.
3. The population of *Halictus* sp. in the two crops varies from 4-10 per cent.
4. *Eristalis* spp. and *Trichometallea pollinosa* Tns. are the only representatives of Diptera that visit *toria* and *sarson* flowers uniformly and regularly. Between themselves they constitute 3-13 per cent of the insect visitors to these flowers.
5. Although the body of *Sepsis* sp. is smooth, but because of its abundance its value as a pollinating agent cannot be doubted.
6. Other insects because of their small numbers, cannot be considered as important pollinators. That they do effect a certain amount of pollination, cannot, however, be doubted.

Hourly frequency of the insect visitors

Observations were made on the hourly frequency of insect visitors to *toria* and *sarson* flowers from 8th December 1931 to 14th December 1931 and from 11th February 1931 to 19th February 1931 daily for 8 hours and 20 minutes. The results of two such observations (one for *toria* and one for *sarson*) are given in Table XX. The method adopted was the same as described by Ali Mohd. *et al.* [1931].

It is seen from Table XX that :—

1. Before 10 a.m. *toria* and *sarson* flowers are mostly visited by Dipterous insects, while after 4 p.m. both Dipterous and Hymenopterous insects are present in almost equal numbers.
2. *Halictus* sp. begins its visits after 9 a.m., *Andrena ilerda* Cam. after 10 a.m., and *Apis florea* Fab. after 11 a.m. From 11 a.m. upto 4 p.m. these three insects completely outnumber all other insect visitors. (These generalizations are confirmed by the remaining 14 observations as well).

SUMMARY

The insect visitors to *toria* and *sarson* flowers were collected for 131 days during November to February in 1930 and 1931, at Lyallpur. This collection includes 105 different species representing 55 families of 9 Orders of the Class Insecta. The habits and usefulness of these insects are discussed and it has been found that *Apis florea* Fab., *Andrena ilerda* Cam., *Halictus* sp. and *Eristalis tenax* Linn. are the most important pollinators.

A study of the hourly frequency of the insect visitors also confirms this conclusion.

Philanthus depredator Smith.—An insect predaceous upon bees—(in its relation to Indian bees) is brought to light for the first time. Its habits and capacity for destruction of bees are described.

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THE HOT FERMENTATION PROCESS FOR COMPOSING TOWN REFUSE AND OTHER WASTE MATERIAL*

III. THE HOT FERMENTATION VS. AEROBIC SYSTEMS OF COMPOSTING

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(With Plate VI)

IT was observed in Part I of this series [Acharya and Subrahmanyam, 1939] that previous studies carried out at Bangalore and at other centres showed that considerable losses of nitrogen and of organic matter occur in the aerobic methods of composting, as ordinarily practised in this country, and that such losses are particularly great while composting street-sweepings with nightsoil. Preliminary trials reported in Part I gave hopes of minimizing the above losses to a considerable extent by adopting the hot fermentation process, wherein the air supply is cut off after five or six days and the mass is allowed to ferment anaerobically for about three months. The various factors which go to control the efficacy of the hot fermentation process have been studied in detail and the results obtained have been reported in Part II [Acharya, 1939]. Having standardized the conditions of the process, it appeared advisable to verify the soundness of the preliminary indications reported in Part I, by carrying out comparative tests of the aerobic and hot fermentation process under controlled conditions on a uniform type of material and on the large scale.

Ragi straw was chosen for the initial trials, with a view to secure uniformity of material and minimize errors in sampling, but in the later experiments street sweepings, screened into the 'organic' and 'soil' fractions were used. Different starters such as ammonium sulphate, cattle dung and urine and nightsoil were tried. The experiments were first carried out in the laboratory in glazed jars, then outdoors in cement-coated concrete cisterns (2 ft. cubes) and finally on the large scale in trenches and overground heaps.

The raw materials used in the experiments had the average chemical composition shown in Table I.

*Parts I and II of this series appeared in *Ind. J. Agric. Sci.* 9, 741—4 and 817—33

TABLE I
Analysis of raw materials used

Constituents	<i>Ragi</i> straw per 100 gm. (sundried)	Cattle dung per 100 gm. (fresh)	Cattle urine per 100 c.c. (diluted)	Nightsoil per 100 gm. (fresh)	Street sweeping screened into	
					Leaves fraction per 100 gm. (air-dry)	Soil fraction per 100 gm. (air-dry)
	gm.	gm.	gm.	gm.	gm.	gm.
1. Dry matter	94.98	20.02	0.809	19.50	90.40	97.60
2. Ash free organic matter .	86.27	15.11	0.359	15.30	54.30	6.20
3. Carbon	38.22	6.60	0.247	9.30	30.71	3.07
4. Nitrogen	0.50	0.278	0.102	1.10	1.23	0.24
5. Ash and non-volatiles .	8.71	4.91	0.450	4.20	36.10	91.80
6. Moisture	5.02	79.98	99.191	80.50	9.60	2.50

JAR EXPERIMENTS

Ragi straw, cut into small bits $\frac{1}{2}$ in. to 1 in. long, was taken up for decomposition, using nightsoil as the starter. Varying quantities of nightsoil necessary to secure different initial C : N ratios of the composts, as shown. in Table II, were weighed into porcelain dishes, the quantities of water shown in the above table were added, the whole well mixed and added in portions to the jars containing *ragi* straw so as to promote uniform admixture. The initial C : N ratios thus obtained were 40 : 1, 30 : 1, 20 : 1 and 15 : 1.

TABLE II
Jar experiments

C : N Ratio (approximate)	<i>Ragi</i> straw taken	Nightsoil added (fresh weight)	Water added
	gm.	gm.	c.c.
40 : 1	200	80	235
30 : 1	200	150	220
20 : 1	200	300	190
15 : 1	200	600	130

One series was allowed to ferment aerobically in shallow wide jars and the samples were turned over once in 10 days and moistened with water, so as to keep the mass moist to the touch, but not to such an extent as to allow any water to accumulate at the bottom of the jar. A parallel series was allowed to ferment by the hot fermentation system. The jars were left undisturbed for six days, at the end of which period the samples were pressed down and covered over with a layer of mud paste and over it a layer of earth. At the end of three months, both series were taken out and the contents dried at 50°—60°C. weighed, powdered and analysed for dry matter, carbon, nitrogen and ash according to the methods followed in Part II of this series.

The percentage composition of the samples obtained in the two series at the end of composting and their C : N ratios, are shown in Table III. It is sometimes the practice to judge the over-all efficiency of a composting process by reference to the analytical composition of the final compost obtained. While this criterion may be the deciding factor in comparing processes, one of which produces a compost of low manurial value (say with a nitrogen content less than 0.5 per cent on dry matter) and another which produces a compost of high manurial value (say, with a nitrogen content of two per cent or over, on dry matter), it cannot be considered to have the same value in cases where both the processes under comparison yield composts above the average quality (i.e. over one per cent nitrogen on dry matter). In such cases, the efficiency of a composting process can best be judged, by taking into consideration the total quantity of manure obtained and the total recovery of manurial constituents such as nitrogen and organic matter secured in the different processes.

The possibility of arriving at misleading inferences by relying solely on the former system of judging the efficiency of a composting procedure will be evident from a reference to Table III. The analytical data presented therein would lead one to infer that the aerobic method of composting is more efficient than the hot fermentation process, since it yields a manure of better quality, containing a higher percentage of nitrogen. In three out of four comparisons (i.e. except at initial C : M ratio of 15 : 1) the nitrogen content of the aerobically prepared manure is definitely higher than that of hot fermented compost. The C : N ratio of the composted material is also narrower in the former case. But as pointed out in the last paragraph, these indications must be judged in conjunction with the total quantities of manure obtained in the two cases and the corresponding recoveries of nitrogen and of organic matter obtained. The importance of this latter consideration will be evident to the farmer who aims at getting the maximum yield of good quality manure from the raw materials he is starting with.

It will be noted from Table III that the quality of the manure obtained by the hot fermentation process is quite satisfactory, since the nitrogen content of the compost is 1.96 per cent on dry matter, even when the initial C : N ratio of the raw material was 40 : 1. The C : N ratio of the final compost varies from 20 : 1 to 10 : 1 depending on the quantity of nightsoil added.

The deciding advantage in favour of the hot fermentation process lies in the much larger amount of manure obtained by that method and the much

higher recoveries of nitrogen and organic matter secured, as compared with the aerobic methods. The relative data bearing on this point are presented in Table IV. In the aerobic methods, the yield of manure (dry weight) obtained is of the order of 30 to 38 per cent, while in the hot fermentation process it ranges from 45 to 52 per cent, i.e. nearly $1\frac{1}{2}$ times as much. A similar relationship reflects itself in the recoveries of organic carbon.

TABLE III

Jar experiments with ragi straw—analysis of composts

Initial C : N ratio of compost lot	Analysis on dry matter			Final C : N ratio of compost
	Ash free organic matter	Carbon	Nitrogen	
<i>Aerobic method</i>	per cent	per cent	per cent	
C : N ratio 40 : 1 . . .	74.99	36.67	2.83	12.96 : 1
Do. 30 : 1 . . .	74.09	39.45	3.03	13.03 : 1
Do. 20 : 1 . . .	68.89	35.40	3.26	10.85 : 1
Do. 15 : 1 . . .	67.39	34.73	3.28	10.60 : 1
<i>Hot fermentation method</i>				
C : N ratio 40 : 1 . . .	82.77	39.45	1.96	20.16 : 1
Do. 30 : 1 . . .	80.97	39.31	2.40	16.35 : 1
Do. 20 : 1 . . .	79.25	36.02	2.87	12.54 : 1
Do. 15 : 1 . . .	75.63	33.62	3.40	9.90 : 1

The economy of nitrogen conservation in the two cases is of particular interest, since nitrogen is the most important manurial constituent present in composts. Where the initial C : N ratio of the compost material is 40 : 1, there is little to choose between the two systems of composting in regard to nitrogen conservation. Both are equally efficient in conserving over 96 per cent of the nitrogen originally present in the raw materials. When the C : N ratio is brought down to 30 : 1 or narrower, the hot fermentation process is found to conserve the original nitrogen better than the aerobic methods. The difference in efficiency between the two methods becomes more marked as the C : N ratio gets narrower.

In interpreting the data contained in Tables III and IV, it must be borne in mind that the raw material used in the present experiments is *ragi* straw, which contains a high percentage of readily decomposable constituents

TABLE IV

Jar Experiments with Ragi straw—Recoveries of Nitrogen and of organic matter during composting

Initial C : N ratio of compost lot	Total dry matter			Ash-free organic matter			Carbon			Nitrogen		
	Taken	Re- covered	Re- covery	Taken	Re- covered	Re- covery	Taken	Re- covered	Re- covery	Taken	Re- cover- ed	Re- covery
	gm.	gm.	per cent	gm.	gm.	per cent	gm.	gm.	per cent	gm.	gm.	per cent
<i>Aerobic method</i>												
C : N ratio 40 : 1 .	216.2	66.8	30.89	199.6	50.1	25.10	77.4	24.5	31.62	1.96	1.89	96.42
Do. 30 : 1 .	232.1	75.3	32.44	211.0	55.8	26.44	84.0	29.7	35.36	2.80	2.28	81.41
Do. 20 : 1 .	266.4	93.5	35.10	237.4	64.4	27.12	97.9	33.1	33.78	4.57	3.05	66.74
Do. 15 : 1 .	335.5	126.4	37.69	289.1	85.2	29.46	125.9	43.9	34.85	8.14	4.14	50.87
<i>Hot fermentation method</i>												
C : N ratio 40 : 1 .	216.2	98.1	45.37	199.6	81.2	40.68	77.4	38.70	50.00	1.96	1.92	97.95
Do. 30 : 1 .	232.1	110.7	47.67	211.0	89.6	42.46	84.0	43.5	51.78	2.80	2.66	94.99
Do. 20 : 1 .	266.4	131.6	49.39	237.4	104.3	43.97	97.9	47.4	48.42	4.57	3.78	82.71
Do. 15 : 1 .	335.5	175.5	52.31	289.1	132.7	45.91	125.9	59.0	46.87	8.14	5.96	73.21

such as hemicelluloses and cellulose. The choice of such a carbonaceous material offers the optimum conditions for maximum conservation of nitrogen in the system under aerobic conditions of composting, since microbial decomposition of the carbonaceous groups is accompanied by a simultaneous stabilization or fixation of the labile nitrogen in the form of microbial bodies. If less readily decomposable materials than straw be used, as is often the case in farming practice, the conservation of nitrogen in the aerobic system may be expected to be less than the figures given in Table IV and the differences between the 'hot fermentation' and 'aerobic' results may be expected to be greater.

It is noteworthy that in the hot fermentation process as much as 82.71 per cent of the nitrogen is conserved even when the initial C : N ratio of the compost lot is as narrow as 20 : 1. This is of particular significance while dealing with the composting of town wastes such as street sweepings and nightsoil. As is evident from the figures given in Table I, the C : N ratio of street sweepings ranges from 20 : 1 to 25 : 1. Street sweepings in India possess generally a narrower C : N ratio than in western countries, on account of the prevailing practice of using leaves as dinner plates in many parts of this country and on account of the considerable amounts of dung and even human excreta which find their way into the sweepings. As such, a mixture of street sweepings and night-soil, under the conditions prevailing in most of our municipalities, has a C : N ratio between 20 : 1 and 15 : 1. The position is aggravated in places where the quantity of sweepings available for composting purposes (especially the organic matter fraction of sweepings) barely exceeds the weight of nightsoil collected, by the municipality. In such cases, very heavy losses of nitrogen may be expected to occur under aerobic methods of composting. The losses could be minimized considerably by adoption of the hot fermentation process.

SEMI-LARGE SCALE EXPERIMENTS WITH *RAGI* STRAW

The jar experiments were followed by experiments on the semi-large scale, using 1000 lb. lots of *ragi* straw and comparing different starters such as minerals, cattle dung and urine and night-soil. The *ragi* straw was cut into bits 6 in. to 8 in. long before use. The mineral starter tried was a mixture of 40 lb. commercial grade ammonium sulphate (92 per cent purity), 50 lb. calcium carbonate (ground chalk) and 5 lb. superphosphate, for every 1000 lb. of straw. The constituents were well mixed and 1/20 portions were sprayed over every 50 lb. of straw added to the compost heap, along with five to six gallons of water to wet the material completely. The quantity of ammonium sulphate added corresponded to about 0.78 per cent nitrogen on the straw, and is approximately near the figure of 0.75 per cent N found to be optimum by Hutchinson and Richards [1921] for the rapid rotting of straw. Calcium carbonate was added to neutralize the acidity produced from ammonium sulphate and super phosphate as well as from the straw. The retentive capacity of straw for water is low, as has been observed by several workers previously, and hence it was found advisable to split up the quantity of water added, by adding 5 gallons per 50 lb. of straw at the beginning and 20 gallons of water to the 1000 lb. lot two or three days afterwards.

In the case of the straw-nightsoil composts, 75 lb. lots of nightsoil (fresh weight) were mixed with five to six gallons of water and added to 100 lb. of straw in portions at a time so that the whole was uniformly mixed. The operations were continued till all the 1000 lb. of straw had been added. It was not necessary to add further water in this case, except in the aerobic methods of composting. The amount of nightsoil added corresponded to 0.825 per cent nitrogen on the straw, which is slightly higher than the 'nitrogen factor' of straw (0.75 per cent N), as found by Hutchinson and Richards [1921] but then the nitrogen in nightsoil is in organic combination and may not be so readily or completely available for microbial utilization as the inorganic salts.

In the third series of experiments, difficulty was experienced in adding cattle dung and urine enough to supply 0.75 per cent of nitrogen on the straw, since dung contained only about 0.27 to 0.28 per cent nitrogen on the fresh weight and the cattle urine available locally was found to be diluted with water and contained only about 0.1 lb. of nitrogen for every ten gallons of urine (Table I). Usually about 20 to 25 lb. of dung are added for every 100 lb. of waste material taken for composting, and the amount of water (or urine) that can be retained by 100 lb. of dry waste material will be roughly about 10 gallons. These amounts of dung and urine will supply only about 0.17 per cent of nitrogen on the straw. In order to increase the quantity of nitrogen added, 100 lb. of dung and ten gallons of urine were added for every 100 lb. of straw. The dung was mixed with the urine in a drum and the straw was added in portions into the drum, stirred well in order to ensure complete wetting with the dung-urine solution and then added to the compost heap. The operations were continued till all the 1000 lb. of straw had been similarly treated. The amount of nitrogen so added by the dung and urine was about 0.38 per cent on the straw,—only half the required amount. It was not found possible to increase this amount further except by adding urine again at later stages, which was not feasible since the hot fermentation heaps were closed anaerobically after six days. Since the mass was sufficiently moist and retained its moisture well, in contradistinction to the case where minerals were added, further additions of urine were not possible even during the above period of six days.

The following gives an outline of the different treatments that were taken up for comparison :—

A. Ragi straw plus minerals (*ammonium sulphate, calcium carbonate and superphosphate*)

- I. Hot Fermentation in bricklined and cemented trenches 4 ft. deep, $3\frac{1}{2}$ feet broad and 10 ft. long.
- II. Hot fermentation in earthen trenches, unbricklined, of the above dimensions.
- III. Aerobic method in shallow wide trenches, 2 ft. deep and 14 ft. square.
- IV. Aerobic method in overground heaps.

B. Ragi straw plus nightsoil—

- V. Hot fermentation in earthen trenches, unbricklined.
- VI. Aerobic method in overground heaps.

C. Ragi straw plus cattle dung and urine—

VII. Hot Fermentation in earthen trenches, unbricklined.

VIII. Aerobic method in overground heaps.

Each trial was carried out in duplicate. In the hot fermentation system, the mass was allowed to decompose aerobically for five to six days. Temperature measurements were made to ensure that there was a satisfactory rise of temperature during this period. In the case of nightsoil composts, the temperature rapidly rose to 65°-70°C. at the end of four days. The dung-urine composts followed up with temperatures of about 60°C. at the end of the above period. In the case of the heaps treated with minerals, the onset of the decomposition was much slower, apparently due to the slowness with which the lignified straw got saturated with the mineral solutions employed. The delay in decomposition might also have been due to the absence of a vigorous microflora in the early stages. Dung and nightsoil are well known to be powerful inocula carrying the necessary organisms for decomposing cellulose and hemicelluloses. The rise in temperature in the compost treated with minerals was, therefore, slower and irregular.

The hot fermented composts, using dung or night soil as starter, were pressed well and covered over with a layer of mud paste two to three in. thick at the end of a week. Over the mud paste cover, a layer of loose earth three to four in. deep was spread, to close any cracks forming in the paste layer below. The hot fermented compost treated with minerals was packed after ten days from the start, since the initial fermentation was slower in this case. The parallel heaps, undergoing aerobic fermentation, were given turnings once in ten days and moistened to the extent necessary to keep the mass moist to the touch. The amount of water so added averaged about 20 gallons per heap at every turning.

At the end of three months, all the compost lots were opened out, carefully discarding the earth at the top of the hot fermentation trenches, and the composts were spread out uniformly on platforms which were bricklined and plastered. It was noticed that whereas the aerobically treated composts had become dark-brown and had broken down to small bits, the hot fermented composts were much lighter in colour and still retained the original shape of the straw pieces. The lighter colour was probably due to the exclusion of air, since the mass rapidly turned dark-brown on exposure to air. The colour change is probably enzymic. As regards texture, the hot fermented composts retained the original shape of the straw since they were not disturbed by being turned over, as in the aerobic method. The material, however, was well decomposed, as shown by the chemical data presented in Table VI and it readily broke down to pieces and was well incorporated in the soil by one ploughing.

After spreading the compost lots uniformly on bricklined platforms, composite samples were taken for analysis by mixing a number of small samples taken at different spots. The residual mass was weighed and stored in closed cisterns for use in field experiments later. The composite sample taken for analysis was weighed, dried at 50-60°C. and again weighed. A portion was taken for moisture determination at 100°C. and the residue was powdered and analysed for carbon, nitrogen, ash and dry matter. From

the values so obtained, the total amounts of dry matter, carbon, nitrogen and ash recovered in the whole quantity of the compost was calculated. The analytical methods used were the same as those adopted in Part I.

The data obtained are presented in Table V. It will be noticed from the above Table that the amount of ash recovered in the case of the cement lined trenches (Treatment I) is almost the same as that originally contained in the compost material at the start, including the added minerals. But in all the other treatments, viz., II to VIII, the amounts of recovered 'ash' are considerably above those present at the start, the difference in some cases going upto 500-600 per cent. of the original value. The difference is smaller in the case of the hot fermentation trenches, where the compost mass has not been disturbed during the process of composting, it is greater in the aerobic method carried out in trenches and is greatest in the aerobic method overground. The increase in apparent 'ash' is no doubt due to admixture of the compost with the surrounding soil and the admixture is greatest in the case of overground heaps which are turned over a number of times. Part of the admixture is brought about during the operation of turning the heap, but a good portion is also brought about by the agency of worms developing in the compost heap.

The large amounts of soil that are thus admixed with the compost are likely to vitiate the inferences drawn from the analytical results, unless due correction be applied for the amount of soil so admixed and the carbon and nitrogen contents of the added soil. Thus the figures given in Table V would lead one to infer that a better recovery of dry matter, representing a larger quantity of manure, is obtained by the aerobic method of composting (Treatments III, IV, VI and VIII), then by the hot fermentation system. Since the organic matter and nitrogen contents of Indian soils are low, the figures given for the recovery of ash-free organic matter, carbon and nitrogen in Table V are vitiated to a smaller extent than those for the recovery of total dry weight of manure.

A correction could be applied for the extraneous soil admixture, by analysing the soil near the locality where the composting is carried out, for its content of ash-free organic matter, carbon, nitrogen and the 'mineral' nonvolatile fraction. By considering the excess of 'ash' recovered in Treatments II to VIII as representing the above 'mineral' fraction of the admixed soil, the corresponding corrections for total dry matter, ash-free organic matter, organic carbon and total nitrogen can be calculated. These values have to be deducted from the corresponding values given in Table V.

The data so corrected for 'admixed soil' are shown in Table VI and present a truer picture of the changes undergone by the raw materials under the different systems of composting. It will be noted therefrom that the hot fermentation system gives a much higher yield of dry matter than the aerobic methods, ranging from $1\frac{1}{2}$ times to twice as much. The recoveries of ash-free organic matter and of carbon by the aerobic method are particularly low in the case of straw (being only 20 to 28 per cent. for heaps overground), since straw is rich in easily oxidizable carbonaceous groups such as the hemicelluloses and cellulose. In such cases, the difference in the relative efficiencies of the hot fermentation and aerobic methods shows itself

in an accentuated form, so far as the recovery of ash-free organic manure is concerned. In the present instance, where nightsoil had been used as the starter, the recovery of ash-free organic matter by the hot fermentation process is over thrice that obtained by the aerobic method. Though similar differences may not be obtained while dealing with more resistant types of waste material such as farm wastes or street sweepings, it is evident that appreciably higher recoveries of organic matter could always be expected from the hot fermentation method.

The conservation of nitrogen also is more efficiently secured in the hot fermentation method than in the aerobic methods, though the relative differences are not of such high magnitude as in the case of organic matter. The conservation is best in the case (Treatment VII) where dung and urine had been used as the starter, but in this case it must be remembered that the amount of nitrogen added in the starter was only about half the 'nitrogen factor' of straw. This would go a great way to explain the very high recovery (92.12 per cent.) of nitrogen obtained in the above treatment. For the same reason, the recovery by the aerobic method also is fairly high (74.71 per cent.) But in cases where the nitrogen added in the starter is equal to or exceeds the 'nitrogen factor' of straw, as in Treatments I to VI the losses in the aerobic methods become greater and the superiority of the hot fermentation process becomes more marked.

Table VII shows the percentage composition and the C/N ratio of the composts prepared by the different methods. While the contents of ash-free organic matter and of carbon are generally higher in the hot fermented composts, the nitrogen percentage is generally lower. This is due to the fact that, though the nitrogen conservation is better in the hot fermentation process the conservation of organic matter is higher still. This is reflected in the C/N ratios of the composts obtained by the two methods. The ratio ranges from 22 to 26 in the case of the hot fermented compost, while it is narrower, viz. from 13 to 15 in the case of aerobically prepared composts. Considering the fact that the initial C : N ratio of *ragi* straw was 76 : 1, the final ratios obtained in the hot fermentation process should be considered to be satisfactory. It is generally agreed that materials with C : N ratios of 20 : 1 and narrower can be safely put on land without showing any adverse effects ; on the other hand, they serve to increase the available nitrogen in the soil (Waksman, 1936). In view of the fact that changing the C : N ratio from 20 : 1 to 12 : 1 or narrower still means, under practical conditions of composting, a simultaneous loss of carbon and nitrogen (Table VI) both of which are of importance in increasing soil fertility, it would obviously be preferable to stop the composting at the C : N ratio near 20 : 1 as in the hot fermentation process, and allow the rest of the decomposition to take place in the soil itself. The practical value and economic advantage of adopting this procedure can only be verified by actual field trials. A detailed account of the field trials that have been conducted with composts prepared by the hot fermentation and aerobic methods will be presented in a later communication, but it may be stated at this stage that the above trials have fully borne out the practical utility of stopping the composting at the earlier stage represented by the hot fermentation process and thus conserving the nitrogen better.

TABLE V
Semi-large scale experiments with ragi straw—recoveries of major constituents
(Uncorrected for soil contamination)

Materials and method of composting	Total dry matter			Ash free organic matter			Carbon			Nitrogen			Ash and Non-volatiles	
	Taken	Re-covered	Recovery	Taken	Re-covered	Recovery	Taken	Re-covered	Recovery	Taken	Re-covered	Recovery	Taken	Re-covered
	lb.	lb.	per cent	lb.	lb.	per cent	lb.	lb.	per cent	lb.	lb.	per cent	lb.	lb.
Ragi straw + Ammonium sulphate														
I. H. F. in bricklined Trench .	990	634	64.04	863	538.8	62.43	382	232.1	60.75	12.81	10.47	81.74	87.0	95.2
II. H. F. in earthen trench un-bricklined.	990	703	71.03	863	474.3	54.96	382	208.6	54.09	12.81	9.52	74.32	87.0	228.7
III. Aerobic in shallow trench .	990	787	79.50	863	306.4	35.50	382	151.4	39.63	12.81	8.06	62.92	87.0	480.6
IV. Aerobic in overground heaps	990	816	82.43	863	215.8	25.00	382	110.8	28.99	12.81	7.29	56.92	87.0	600.2
Ragi straw + Nightsoil														
V. H. F. in earthen trench .	1,100	925	84.09	978	618.3	63.23	451.8	264.1	58.45	13.25	11.74	88.61	130.9	306.7
VI. Aerobic in overground heaps	1,100	976	88.72	978	199.4	20.39	451.8	98.1	21.72	13.25	7.69	58.03	118.5	776.6
Ragi straw + Dung + Urine														
VII. H. F. in earthen trench .	1,168	796	68.74	1,018	515.1	50.62	450.5	214.9	47.69	8.78	8.16	92.94	140.5	280.9
VIII. Aerobic in overground heaps	1,168	934	80.65	1,018	221.5	21.74	450.5	92.4	20.51	8.78	6.85	78.00	140.5	712.5

TABLE VI

Semi-large scale experiments with ragi straw—recoveries of constituents after correction for soil contamination

Materials and method of composting	Total dry matter			Ash free organic matter			Carbon			Nitrogen		
	Taken	Recovered	Recovery	Taken	Recovered	Recovery	Taken	Recovered	Recovery	Taken	Recovered	Recovery
	lb.	lb.	per cent	lb.	lb.	per cent	lb.	lb.	per cent	lb.	lb.	per cent
Ragi straw + Am. sulphate												
I. H. F. in bricklined trench .	990	634	64.04	863	538.8	62.43	382	232.1	60.75	12.81	10.47	81.74
II. H. F. in earthen trench un-bricklined.	990	561	56.67	863	472.8	54.78	382	205.7	53.86	12.81	9.45	73.77
• III. Aerobic in shallow trench .	990	398	39.70	863	302.0	34.99	382	149.0	39.00	12.81	7.86	61.35
IV. Aerobic in heaps overground	990	303	30.61	863	210.2	24.35	382	107.6	28.17	12.81	7.04	54.96
Ragi straw + Nightsoil												
V. H. F. in earthen trench .	1,100	750	68.19	978	616.4	63.04	451.8	243.0	58.21	13.25	11.65	87.94
VI. Aerobic in heaps overground	1,100	318	28.91	978	192.3	19.66	451.8	94.1	20.83	13.25	7.36	55.55
Ragi straw + Dung + Urine												
VII. H. F. in earthen trench .	1,168	654	56.48	1,018	513.6	50.47	450.5	214.0	47.50	8.78	8.09	92.12
VIII. Aerobic in heaps overground	1,168	356	30.74	1,018	215.2	21.14	450.5	88.8	19.71	8.78	6.56	74.71

TABLE VII

Semi-large scale experiments with ragi straw—analysis and C/N ratio of composts

Materials and method of composting	Initial C/N ratio of compost lot	Analysis on dry material			Final C/N ratio of compost
		Ash free organic matter	Carbon	Nitrogen	
<i>Ragi straw + Am. sulphate</i>		per cent.	per cent.	per cent.	
I. H. F. in bricklined trench.	29.83 : 1	84.98	36.61	1.651	22.17 : 1
II. H. F. in earthen trench unbricklined.	29.83 : 1	84.26	36.67	1.685	21.77 : 1
III. Aerobic in shallow trench.	29.83 : 1	76.85	37.91	2.000	18.95 : 1
IV. Aerobic in heaps over-ground.	29.83 : 1	69.37	35.52	2.324	15.29 : 1
<i>Ragi straw + Nightsoil</i>					
V. H. F. in earthen trench.	34.10 : 1	82.19	35.06	1.553	22.57 : 1
VI. Aerobic in heaps over-ground.	34.10 : 1	60.47	29.59	2.315	12.78 : 1
<i>Ragi straw + Dung + Urine</i>					
VII. H. F. in earthen trench.	51.31 : 1	78.52	32.72	1.237	26.45 : 1
VIII. Aerobic in heaps over-ground.	51.31 : 1	60.45	24.95	1.843	13.54 : 1

CISTERN EXPERIMENTS WITH TOWN REFUSE

As stated in an earlier paragraph, straw is a highly carbonaceous material which shows in an accentuated form the differences between the two systems of composting under comparison, and it seemed therefore advisable to repeat the experiments using a more resistant material such as town refuse, in place of *ragi* straw. In order to secure a certain degree of uniformity in the lots taken for the different comparisons, the sweepings were sieved through an expanded metal sieve set at 5/8 in., into a fraction, which consisted mostly of soil and ash, and another fraction consisting mostly of leaves, paper and other organic materials. Under Indian conditions, where there is plenty of vegetation in towns, the second fraction consists mostly of leaves. For

convenience of nomenclature, the two fractions into which the street sweepings are sieved are designated respectively as the 'soil' and 'leaves' fractions. The average chemical composition of the two fractions is shown in (Table 1).

The experiments were carried out in the first season in cement-coated concrete cisterns 2 ft. \times 2 ft. \times 2 ft. in dimensions. Nightsoil and cattle dung *plus* urine were compared as starters. Since in some municipalities there is a custom of sieving out the 'organic matter' fraction of street sweepings and using that alone for composting purposes and other municipalities use a mixture of both fractions, it seemed advisable to compare both systems. It has been already reported in Part II of this series [Acharya, 1939] that the addition of moderate quantities of the soil fraction of street sweepings exerted a beneficial effect on the course of the decomposition and on nitrogen conservation and that a quantity of soil corresponding to half the weight of night soil taken for composting appeared to be optimum.

(i) *Cistern experiments with street sweepings and nightsoil*

In the series where nightsoil was added as the starter, 100 lb. lots of the 'leaves' fraction of street sweepings, with and without the addition of 50 lb. of the 'soil' fraction, were uniformly mixed in portions inside the cisterns, with 100 lb. of nightsoil diluted with five to six gallons of water. The following treatments were compared using duplicate samples—

(a) aerobic decomposition throughout, with the material turned over once in 10 days and moistened when necessary ; (b) hot fermentation process wherein the material was covered over with a layer of mud paste and over it earth, after a preliminary period of six days aerobic fermentation ; and (c) anaerobic decomposition from the start, by packing the material well and covering it with mud paste and a layer of earth from the beginning.

At the end of three months, the residual material was taken out, spread on a bricklined and plastered platform, weighed and samples taken for analysis. The sampling and analytical procedures were the same as those described earlier in this paper in Section II for the composting of *ragi* straw on the large scale.

The analytical data obtained are summarised in Table VIII. Since the experiments were carried out in cisterns and the soil covering at the top was removed carefully, there was no appreciable admixture with extraneous soil and hence no correction for 'soil contamination' was necessary as was applied in the case of the *ragi* straw experiments on the large scale described in Section II of this paper. The data presented in Table VIII confirm the previous conclusions that the hot fermentation method gives much higher recoveries of ash-free organic matter and of nitrogen than the aerobic method. In contradistinction to the experiments on *ragi* straw (Table VII), the percentage of nitrogen in the hot fermented compost is higher in the present experiment than the percentage in aerobically prepared composts. This is due to the fact that *ragi* straw is poor in nitrogen (0.5 per cent. N) and in the experiments described in Section II above only about 0.825 per cent of nitrogen was added to the straw in the form of nightsoil. The initial C : N ratio of the straw-nightsoil mixture was nearly 34 : 1. A reference to the jar experiments described in Section I above (Table III) would show that at C : N ratios

between 30 : 1 and 40 : 1, the aerobically prepared compost shows a higher percentage of nitrogen than the hot fermented compost, even though the total recovery of nitrogen by the former method is somewhat lower than that obtained by the latter method.

In the present cistern experiments, however, the organic fraction of street sweepings have been used in place of *ragi* straw and a reference to Table I would show that these are richer in nitrogen (1.23 per cent). The initial C : N ratio of the mixture of sweepings and nightsoil was about 18 : 1, which is much narrower than the ratio of 34 : 1 obtained for straw : nightsoil mixtures. A reference to Tables III and IV would show that at such narrow C : N ratios, the loss of nitrogen in the aerobic method is very high and is greater than the loss in carbon. Hence, under such conditions, the hot fermented composts show a higher percentage of nitrogen, along with a higher yield of manure than by the aerobic system. Under the conditions existing in Indian towns and villages, where the street sweepings contain a large amount of leaves and animal excreta, it is doubtful whether it would be possible to widen the C : N ratio of the mixture of sweepings and nightsoil beyond 20 : 1 at the start. Under these conditions, the losses of nitrogen by the aerobic methods are bound to be heavy.

TABLE VIII
Cistern experiments with town refuse—nightsoil as starter

Without soil 100 lb. leaves fraction of street sweepings. 100 lb. Nightsoil	Yield of manure		Carbon		Nitrogen		Ash free organic matter	
	Fresh	Sundry	On sundry material	Recovery	On sundry material	Recovery	On sundry material	Recovery
	lb.	lb.	per cent	per cent	per cent	per cent	per cent	per cent
1. Aerobic Method . .	126	68	23.24	39.50	1.37	39.97	38.18	37.30
2. Hot Fermentation Process.	181	85	25.30	53.74	1.62	59.09	45.45	55.51
3. Anaerobic from the start.	202	92	31.57	72.59	1.79	70.68	54.09	71.50
<i>Added soil.</i>								
100 lb. leaves fraction of street sweepings. 50 lb. Soil fraction . 100 lb. Nightsoil.								
1. Aerobic Method .	252	137	15.1	49.80	0.76	42.44	22.0	41.47
2. Hot Fermentation Process.	334	149	19.1	68.52	1.18	71.76	30.5	62.52
3. Anaerobic from the start.	371	159	20.8	79.64	1.28	82.07	33.2	72.61

It will be noticed from Table VIII that the material which was anaerobically packed from the very start gave the highest recovery of nitrogen and of organic matter, and it may appear as though this method would be an improvement over both the aerobic and hot fermentation methods. But the method of anaerobic packing possessed several disadvantages in practice ; e.g. the system

did not promote the development of an active microflora which would decompose nightsoil, as evidenced by the fact that when the cisterns were opened out at the end of three months, the material smelt strongly of nightsoil, and undecomposed masses of nightsoil could be seen. Nitrification experiments in the laboratory, to be described in a subsequent communication, showed that a much longer initial lag period was required by the anærobically prepared composts before nitrification set in, than in the case of the hot fermented composts. This initial lag period was probably due to the presence of undecomposed carbonaceous material of the straw. Field trials with such anærobically prepared composts, to be presented in a future communication, showed that the crop response was poorer in the first season, when compared to the hot fermented composts, but was equal to them in the second season. These facts would indicate that a purely anærobic method of composting either fails to remove substances or causes the formation of substances which interfere with the nitrification processes in the soil and with plant growth. It would, therefore, seem advisable to ensure a suitable combination of the aerobic and anærobic treatments, as is done in the hot fermentation method.

It will also be noted from Table VIII that the addition of moderate amounts of the soil fraction of street sweepings improves the conservation of organic matter and to a greater extent of nitrogen. The compost appeared to be better broken down in the presence of soil. The addition of soil, however, lowers the percentage of nitrogen and of organic matter in the compost and, as already observed in Part II, it would be advisable to limit the addition of soil to the minimum quantity necessary to promote the better decay of the compost material. The proportion used in the present experiments of 2 : 1 : 2 (by weight) between the leaves fraction, the soil fraction and nightsoil would appear to work satisfactorily.

(ii) *Cistern experiments with street sweepings and cattle dung plus urine*

Another series of compost experiments in cisterns was run on the same lines as the previous series described in Section III (i) above, but with the difference that a mixture of cattle dung and urine was used as the starter instead of nightsoil. A certain amount of household (wood) ash was also added to correspond with the conditions in farming practice. For every 100 lb. of leaf material, with and without the addition of 50 lb. of soil, 25 lb. of dung and three gallons of urine were added. Extra water to the extent of eight to nine gallons was also added. The treatments compared were the same as in Section III (i), viz. aerobic, hot fermentation and completely anærobic, and also the influence of the addition of soil. The details of composting procedure and methods of sampling and analysis were the same as described in Section III (i).

The analytical results obtained are summarized in Table IX. The general trend of the results is the same as in the case of the previous experiments with nightsoil, viz., the hot fermentation method secures a better conservation of organic matter, carbon and nitrogen than the aerobic method, both as regards the percentage of these constituents present in the manure as well as in the absolute amounts recovered out of the quantities originally present in the raw materials. The anærobically prepared composts contain

a higher percentage of nitrogen, but at the same time they contain large amounts of readily oxidizable constituents of the original raw material and hence are open to the objections and drawbacks referred to in the last section III (i) while dealing with refuse-nightsoil composts. The protective action of the soil in lessening the loss of nitrogen and of organic matter may be noticed in this case also.

TABLE IX

Cistern experiments with town refuse—cattle urine and dung as starters

Without soil 100 lb. leaves. 10 lb. Household ash. 25 lb. dung and 3 gallons cattle urine.	Yield of manure		Carbon		Nitrogen		Ash free organic matter.	
	Fresh	Sundry	Sundry material	Recovery	Sundry material	Recovery	Sundry material	Recovery
	lb.	lb.	per cent	per cent	per cent	per cent	per cent	per cent
1. Aerobic method	109	67	18.2	39.32	0.72	38.59	27.0	32.96
2. Hot fermentation process.	178	80	21.5	55.46	1.06	67.84	36.2	52.74
3. Anaerobic from the start.	201	89	25.3	72.61	1.18	84.09	41.5	67.26
Added soil 50 lb. soil. 100 lb. leaves. 10 lb. Household ash. 25 lb. dung and 3 gallons urine.								
	Fresh	Sundry	Sundry material	Recovery	Sundry material	Recovery	Sundry material	Recovery
	lb.	lb.	per cent	per cent	per cent	per cent	per cent	per cent
1. Aerobic method	196	112	16.4	56.44	0.70	57.23	28.2	54.45
2. Hot fermentation process.	271	125	20.2	77.52	0.86	78.48	34.2	73.70
3. Anaerobic from the start.	284	134	21.4	88.12	0.90	88.02	37.8	87.34

A comparison of Tables VIII and IX would show that the composts prepared by use of cattle dung and urine are appreciably poorer in nitrogen as compared with those prepared by use of nightsoil as starter. This is due to the fact that cattle dung contains only about a fourth of the nitrogen contained in nightsoil (Table I) and the quantity of cattle urine that can be added to the compost heap is limited by the capacity for absorption by the raw materials. These difficulties have been already referred to in detail in Section II above. The difference in nitrogen contents of the two starters is reflected also in the manurial values of the respective composts when they are applied to land. Nightsoil composts, if they are properly prepared, give much better crop responses, per ton of manure, than composts prepared by use of dung and urine, as has been shown by field experiments which will be reported in a further communication.

SEMI-LARGE SCALE EXPERIMENTS WITH TOWN REFUSE

The cistern experiments with town refuse were followed up by semi-large experiments, using lots of 1,000 lb. of the sieved 'leaves' fraction of street

sweepings and 500 lb. of the 'soil' fraction. In the present series of experiments an effort was made to see whether the capital cost of digging the trenches could be avoided by carrying out the hot fermentation process in overground heaps, by mud-plastering these heaps after an initial aerobic fermentation of six to seven days, and to see how such composts compare with those prepared in trenches. The following methods of composting were compared :—

- (1) Hot Fermentation in underground trenches which were bricklined and plastered.
- (2) Hot Fermentation in underground trenches without bricklining.
- (3) Hot Fermentation method in heaps overground on bricklined platforms.
- (4) Hot Fermentation method in heaps overground on platforms which were not bricklined.
- (5) Aerobic process in shallow wide trenches which were not bricklined.
- (6) Aerobic process in heaps overground on platforms which were not bricklined.

Two different starters were tried, viz. nightsoil and cattle dung *plus* urine. Each trial was carried out in duplicate.

In the first series using nightsoil as starter, lots of 100 lb. of 'leaves' fraction of street sweepings and 50 lb. of the 'soil' fraction were uniformly mixed within the container itself with 100 lb. of nightsoil (fresh weight) diluted with eight to nine gallons of water. The operations were repeated ten times, so that in all 1,000 lb. of 'leaves' fraction and 500 lb. of 'soil' fraction and 1,000 lb. of nightsoil were added to form a compost lot. In the parallel series using cattle dung *plus* urine as starter, 25 lb. of dung and five gallons of urine were mixed with further ten gallons of water and uniformly mixed with lots of 100 lb. of 'leaves' fraction and 50 lb. of 'soil' fraction. The operations were repeated till 1000 lb. of the 'leaves' fraction and 500 lb. of 'soil' fraction had been so treated. The moisture level was brought to about 50 per cent. at the start.

The hot fermentation composts, either in trenches or in heaps overground, were packed free from air after six days, by a layer of mud plaster and earth, as already described in section II of this paper. The 'aerobic' composts were turned over once in 15 days with watering, enough to keep the mass moist to the touch. The total amount of water so added ranged from 25 gallons per heap per turning in the initial stages to about 10 gallons towards the final stages of decomposition.

At the end of three months, the composts were taken out, spread uniformly on bricklined platforms, samples taken for analysis, the residual mass weighed, and the samples analysed according to the procedure already outlined for the semi-large scale experiments with *ragi* straw in section II above. In the present case also, it was noticed that there was a large admixture of extraneous soil in the composts prepared overground on unbricklined platforms, and to a lesser extent in trenches which were not bricklined. The contamination was still less in the case of heaps kept on bricklined platforms or in bricklined trenches. Necessary corrections have been applied for the above soil contamination, according to the method described in Section II above, and the corrected

data are presented in Tables X and XI. Table X gives the results obtained by use of nightsoil as starter and Table XI gives the corresponding data for composts prepared by use of cattle dung *plus* urine as starter.

TABLE X

Semi-large scale experiments with town refuse using nightsoil as starter.

Taken— 1,000 lb. leaf fraction of street sweepings. 500 lb. soil fraction of street sweepings. 1,000 lb. Nightsoil (fresh).	Yield of manure		Carbon		Nitrogen		Ash free organic matter	
	Fresh	Sun-dried	Analysis	Recovery	Analysis	Recovery	Analysis	Recovery
	lb.	lb.	per cent	per cent	per cent	per cent	per cent	per cent
I. Hot Fermentation in underground trenches with brick-lining.	3,215	1,520	19.81	72.46	1.22	75.91	34.93	73.05
II. Hot Fermentation in underground trenches without bricklining.	3,010	1,438	18.01	62.34	1.13	66.36	31.85	63.01
III. Hot Fermentation in heaps overground on bricklined platforms.	2,760	1,325	17.84	56.90	1.19	64.34	30.39	55.39
IV. Hot Fermentation in heaps overground on platforms not bricklined.	2,820	1,280	17.05	52.53	1.10	57.58	29.55	52.02
V. Aerobic, in shallow wide trenches.	2,230	1,185	16.60	47.36	1.08	52.49	29.36	47.87
VI. Aerobic, in heaps over-ground.	1,980	1,042	14.98	37.58	0.99	42.09	28.89	41.41

TABLE XI

Semi-large scale experiments with town refuse using cattle urine and dung as starters

Taken— 1,000 lb. leaf fraction of street sweepings. 500 lb. soil fraction of street sweepings. 250 lb. dung. 50 gallons of urine.	Yield of manure		Carbon		Nitrogen		Ash free organic matter	
	Fresh	Sun-dry.	Analysis	Recovery	Analysis	Recovery	Analysis	Recovery
	lb.	lb.	per cent	per cent	per cent	per cent	per cent	per cent
I. Hot Fermentation in underground trenches with brick-lining.	3,015	1,415	17.81	77.45	0.77	79.52	31.11	75.88
II. Hot Fermentation in underground trenches without bricklining.	2,784	1,320	17.04	69.15	0.74	71.53	28.78	65.52
III. Hot Fermentation in heaps overground on bricklined platforms.	2,645	1,275	16.47	64.54	0.69	64.12	26.27	57.76
IV. Hot Fermentation in heaps overground on platforms not bricklined.	2,496	1,190	15.13	55.32	0.63	54.75	25.66	52.64
V. Aerobic, in shallow wide trenches.	2,112	1,075	13.95	46.10	0.60	47.10	24.82	46.03
VI. Aerobic, in heaps overground	1,826	960	13.02	38.41	0.59	41.35	23.44	38.80



FIG. 1. A battery of cisterns in which the preliminary studies were carried out under controlled conditions



FIG. 2. Composting of night soil with street sweepings in trenches

It has been mentioned above that one of the objects of the present experiment was to see whether trenches could be dispensed with in the hot fermentation method and overground heaps used instead. The data presented in Tables X and XI reveal that in the case of refuse-nightsoil composts as well as refuse-dung composts, trenches gave better results than overground heaps. This was shown both in the hot fermented composts and in those aerobically prepared. The superiority of trenches to overground heaps in conserving a greater amount of nitrogen and of organic matter, has been already noticed by Aiyar [1933] and others and is attributable to the more efficient retention of moisture and the more rapid rise of temperature secured in trenches. The temperature is also maintained at a high level for a longer time in trenches. Waksman and coworkers [1939] have found that the loss of nitrogen during composting is less in cases where there is a rapid rise of temperature in the early stages; the loss was greatest in cases where the temperature rise is slow and irregular.

That the better conservation of nitrogen and of organic matter in the hot fermentation method is not solely due to the composting being carried on in trenches in that method, is shown by comparing the results obtained (Tables X and XI) for the treatments II and IV, where both the hot fermentation and aerobic methods are carried out in trenches, and also treatments IV and VI where both the above methods have been tried on heaps overground. In both cases, the hot fermentation system has given a higher recovery of organic matter and of nitrogen, indicating that in the aerobic method, as ordinarily practised, there is a continuous loss of nitrogen, which could be avoided by packing the material anaerobically after the initial stage of aerobic fermentation.

A comparison of the data shown in Table X with those given in Table XI confirm the observations made in Section III (ii) above, regarding the relative manurial values of composts prepared from nightsoil and cattle-dung as starters. In the present series also, the nightsoil composts show appreciably higher percentages of nitrogen and of ash-free organic matter than those prepared from dung and urine.

A comparison of treatments II and VI (Tables X and XI) would show that by adopting the hot fermentation system in trenches, it is possible to obtain recoveries of nitrogen and of organic matter which are about $1\frac{1}{2}$ times those obtained by the aerobic method overground.

DISCUSSION

It is generally admitted that nitrogen is the most important constituent present in bulky organic manures like composts, next to the organic matter contained. Most Indian soils being deficient in phosphoric acid, the amount of this constituent also may go to a certain extent in influencing the manurial value, especially in the case of nightsoil composts which are rich in P_2O_5 . But there is little likelihood of loss of phosphoric acid during the process of composting, if losses due to leaching be avoided, whereas losses of nitrogen occur often to a considerable extent and are not easily avoided.

The experiments described above have shown that the loss of nitrogen is particularly serious while dealing with the disposal of town wastes such as

nightsoil and street sweepings by the method of composting, on account of the narrow C : N ratio of the materials concerned. When dealing with materials of such narrow C : N ratio, the aerobic methods involve heavy losses of nitrogen to the extent of 50 per cent and more of the quantity originally present. The loss of nitrogen implies a proportionate decrease in the manurial value of the compost and hence in the price which the manure could obtain. Conversely, by adopting a method of composting which would minimize the loss, it would be possible to prepare a correspondingly larger quantity of manure containing the same percentage of nitrogen. By either alternative, it will be possible for a municipality or a private farmer to obtain a greater return from the available raw materials on hand. Hence arises the need for devising a method which, under practical conditions of composting, would minimize the loss of nitrogen, if not avoid it altogether. Since Indian soils are poor in organic matter, it would at the same time be an advantage if more of the organic matter could be conserved than what is done by the aerobic methods. Judged by these two tests, the hot fermentation method appears to be a distinct improvement over the ordinary aerobic methods of composting. The data given in the present paper would clearly show that in the aerobic methods of composting, especially under our Indian conditions of high temperatures and in some areas excessive rainfall, the oxidation is carried to an extreme stage, when heavy and avoidable losses of nitrogen and of organic matter occur. It would obviously be an advantage to stop the aerobic decomposition at a much earlier stage, as is done in the hot fermentation method, and allow the rest of the decomposition to proceed anaerobically.

Two seeming advantages of the aerobic method of composting, which are often quoted, are : (1) the rapidity with which the composting is finished and the material is ready for application to land ; and (2) the thorough decomposition of the material to a dark-brown, friable powder which could be easily incorporated in the soil. But the soundness of these arguments still needs experimental proof. As regards argument No. 1 above, viz. the rapidity of composting, it is of advantage only in some cases, e.g. in the disposal of town refuse by municipalities. But if the process involves simultaneously heavy losses of nitrogen, the financial loss involved thereby is a factor which cannot be ignored. In the case of the private farmer, the rapidity of decomposition may prove a serious disadvantage, since the farmer requires his manure only once in six months and in many areas only once a year for application to land. He will be faced with the problem of how best to store the manure without loss of manurial value, if the composting be finished several months in advance of the date when he may be requiring it.

As regards argument No. 2 above, viz. the degree of decomposition to which a material should be subjected before it is fit for application to land, this is a question on which divergent views are held and convincing experimental evidence is lacking. There is no experimental evidence to show that it is necessary to decompose a material to the ultimate stage of a dark-brown, friable powder before it is fit for application to land. The earlier experiments regarding the unsuitability of straw for direct application to land have been found to be due to the wide C : N ratio of such material, and composting has been found to narrow the above ratio. It has been claimed that soil organic matter has

an approximate C : N ratio of 10 : 1 and that aerobically prepared composts finally approach this ratio and hence are in a fit condition to be applied to land. Waksman in his book on 'Humus' points out that materials of C : N ratio near 15 : 1 do not immobilize soil nitrogen but tend to increase the available nitrogen in the soil. The above ratio is apparently much wider than the ratio of 10 : 1 aimed at in the aerobic methods of composting.

Field experiments have been proceeding at Rothamsted (England) for some years past wherein the compost prepared by the Adco method with a C : N ratio near 11 : 1 has been compared with the direct application to the land of straw and the minerals used for compost preparation, the C : N ratio of the uncomposted materials being near 30 : 1. The levels of nitrogen, phosphoric acid and potash applied in the two cases are the same. The experiments have been proceeding only for the last five or six years, but the results obtained so far show that the uncomposted materials give as good crop responses as the compost.

The above would show that the C : N ratio of a compost material is not the only criterion for judging the suitability of its application to land. On the other hand, a better criterion would be the presence of readily available nitrogen in the material enough to supply the 'nitrogen factor' or the nitrogen requirements of the easily decomposable fractions in the raw material. If for financial or other reasons it is not found advisable to add mineral supplements in order to increase the available nitrogen in the material, the process of fermentation (or composting) has to be resorted to in order to remove the readily oxidizable constituents of the raw material. This should be carried out to such an extent as not to lose any appreciable quantity of the nitrogen originally present in the raw material. Such losses usually fall on the readily available groups such as ammoniacal or nitrate nitrogen and hence greatly lower the manurial value of the product when put on the field. Considered in this light, the hot fermentation method not only conserves a greater portion of the original nitrogen, but also minimizes the loss of the best portion of it, from the manurial point of view.

The question whether composting could be avoided altogether and the raw materials can be directly put on the land requires further and large scale experimentation before a definite answer could be given. The Rothamsted experiments indicate that under certain conditions, it is possible to do so, especially when the raw materials are supplemented by suitable quantities of nitrogenous artificials. Somewhat similar results have been obtained by the Tocklai Experimental Station in some of their experiments on tea*. But the experiments need repetition at a number of other centres in India where climatic and soil conditions and crops raised are different. In both the Rothamsted and Tocklai experiments, water supply has not been the limiting factor, and it is well known that with satisfactory water supply, partially decomposed materials can be safely put on land. Unfortunately, water supply is a serious limiting factor in most areas of this country.

It would also be interesting to know whether the Rothamsted results with straw could be duplicated in this country, using cattle dung and urine as the nitrogenous supplement to straw in place of artificials. The availability of

* Private communication,

nitrogen in the dung *plus* urine starter would be lower than in artificials and it would be useful to know whether with such a supplement the undecomposed raw materials when directly put on the land would prove beneficial to plant growth. In such cases it may be found necessary to remove the readily oxidizable constituents of the raw materials by a preliminary process of fermentation, at least to the extent secured in the hot fermentation process.

While our knowledge of the conditions under which undecomposed materials could be safely put on land to serve as manure for a crop already on the land or about to be sown in is still indefinite, the field trials that have been carried on with hot fermented manures have shown that they can be safely applied to land at any time, with beneficial results.

In addition to the larger recovery of manure and of nitrogen obtained by the hot fermentation method, this method possesses certain important advantages to the farmer from the practical point of view, namely in relation to labour and water requirements. The question of water requirement has been dealt with in detail in an earlier communication [Acharya, 1939] and it was shown therein that the hot fermentation process requires only about a third of the water supply required by the aerobic method, since the mass is packed anaerobically after six days in the former method and no further additions of water are made. Water supply is a serious and in some cases a costly problem in vast areas of this country, e.g. in the Central Provinces, Deccan, etc. In such areas the hot fermentation method would prove particularly useful.

Secondly, the hot fermentation process requires much less labour than the aerobic method, since in the former case there are no periodical turnings and waterings to be given. No attention has to be paid to the compost after once it is packed in the trenches and closed up after a week, till the compost is actually required for application to land. As such, the farmer obtains the compost at a much lower cost and with much lesser trouble than in the case of the aerobically prepared compost. Estimates show that the labour charges for making one ton of compost by the hot fermentation method comes to about 8 As., when working on the large scale, whereas in the aerobic method the charges amount to about three or four times as much.

The hot fermentation system of composting, therefore, appears to be peculiarly suited to the conditions existing in this country.

SUMMARY AND CONCLUSIONS

A comparison of the hot fermentation and aerobic systems of composting was made under controlled conditions: (I) with *ragi* straw in jars on a laboratory scale; (II) with *ragi* straw on the large scale in trenches and overground heaps; (III) with town refuse in concrete cisterns; and (IV) with town refuse on the large scale in trenches and overground heaps. Nightsoil, cattle dung *plus* urine and a mixture of mineral salts consisting of ammonium sulphate, calcium carbonate and superphosphate were used as starters for composting. Different modifications of the aerobic and hot fermentation methods, such as carrying out the composting in trenches and in overground heaps were tried. In the aerobic method, the heaps were turned over once in 15 days and watered to the necessary extent. In the hot fermentation method, the compost lots, generally in trenches, were allowed to ferment aerobically for six

days and then covered with a layer of mud paste and over it with loose earth. The comparison limited itself mainly to the efficiency of conservation of nitrogen, carbon and organic matter, since minerals such as calcium, potash and phosphates are not lost to a great extent, if the composting process be carried out with care.

The experimental data obtained showed :—

1. In all the cases examined, the hot fermentation method gave a much higher yield of manure, as measured by the organic matter contained, than the aerobic method, the ratio varying from $1\frac{1}{2}$ times to 3 times. In cases where the composting is carried out on plain ground or in unbricklined trenches, there is heavy contamination with extraneous soil, for which due correction should be made in the analytical figures.

2. The hot fermentation method conserved the original nitrogen of the refuse materials, in all the cases, examined to a greater extent than the aerobic method. The loss of nitrogen in the aerobic method was particularly great when nightsoil was composted with street sweepings and in some cases amounted to 50 per cent or more of the original amount.

3. The percentages of carbon and of organic matter in the final compost (after making a correction for the extraneous soil contamination) were also higher in the hot fermented composts than in the aerobically prepared composts.

4. As regards the relative percentages of nitrogen in the above two classes of composts, this varied with the initial C : N ratio of the refuse materials. Where the initial C : N ratio was wider than 30 : 1, the increase in conservation of organic matter by the hot fermentation process over the aerobic method was greater than the increase in conservation of nitrogen, with the result that the final percentage of nitrogen in the compost is lower in the hot fermentation method, though the total quantity of nitrogen recovered is greater in that method than in the aerobic. Where the initial C : N ratio is narrower than 30 : 1, the hot fermented composts possess a higher percentage of nitrogen in the final compost, along with a larger recovery of nitrogen in the total compost obtained.

5. The C : N ratio of the compost obtained by the hot fermentation method varies from 15 : 1 to 20 : 1, while that obtained by the aerobic method ranges from 11 : 1 to 15 : 1.

6. Carrying out the composting in trenches ensures a higher yield of manure and a better conservation of nitrogen than the use of overground heaps for composting.

7. Composts prepared by use of nightsoil, were generally richer in nitrogen and of better quality than composts prepared by use of cattle dung and urine as starters.

8. The addition of moderate quantities of earth promoted better decay of the compost and a better conservation of nitrogen, especially in cases where nitrogen-rich materials such as nightsoil were being composted.

9. The hot fermentation process requires only about one half to one third the water supply required by the aerobic methods and appears to be particularly suited to the 'dry' areas of the country.

10. Labour charges in the hot fermentation process are only about a third or fourth of those necessary to carry out the aerobic method of composting, and the attention and supervision required in the former case are considerably less than in the latter case.

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IV. THE HOT FERMENTATION vs. POUDRETTE METHODS FOR THE DISPOSAL OF NIGHTSOIL

BY

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IN a previous communication in this series [Acharya, 1940] the relative advantages of the hot fermentation and aerobic methods of composting were discussed, with special reference to the treatment of town wastes rich in nitrogen, such as nightsoil, and it was shown therein that a better conservation of nitrogen and organic matter was secured in the former method as compared with the latter. Very few municipalities, however, adopt at the present day either of the above systems of composting for the disposal of their nightsoil in spite of active propaganda in this direction by some of the Departments of Agriculture in India—especially those in Madras and Bombay. The method generally adopted by over 90 per cent of the municipalities in this country is that known as ‘pitting’ or ‘trenching’ nightsoil, carried out at nightsoil depots situated at some distance outside municipal limits. In this method, the nightsoil is let out into trenches three or four feet broad and equally deep, dug in the ground, and is then covered over with a layer of rubbish or more usually with earth. The Nasik Municipality has paid particular attention to the details of a proper system of trenching so as to avoid odours and flybreeding and to hasten the drying up of the material. The product so dried is usually designated as ‘poudrette’ and sold as manure to the ryots.

The ‘poudrette’ method of disposal of nightsoil has been in general use in municipalities in this country for the last five or six decades and is even now preferred to the method of ‘composting’, since it is felt that the former method is simpler to carry out and requires less expense and supervision. But, unfortunately, this method of disposal brings little return to the municipalities. The annual income realized by sale of the poudrette in the trenches after a year of drying, amounts on the average to a few hundred rupees only as against the expense of several thousands per year incurred for the collection of nightsoil and street rubbish. This low income is partly attributed to the persistence of the smell of nightsoil in the poudrette, since the latter is more or less only dried nightsoil, and it is well known that the Indian ryot has a sentimental aversion to the handling of such a product [Leather, 1895]. It is also possible that during the process of poudrette-making, as ordinarily practised by municipalities, a good portion of the manurial value is lost from the nightsoil as may be inferred from a typical analysis quoted by Leather [1895] of a poudrette prepared in Cawnpore, viz.

TABLE I

Analysis of poudrette from Cawnpore

	Percentage
Moisture . . .	2.64
Organic matter .	7.82
Earthy substance .	89.54
Nitrogen . . .	0.468
Phosphoric acid .	0.499

In view of the prevalent custom of most municipalities in India, of converting their nightsoil into poudrette, it was considered advisable to compare the relative efficiencies of the poudrette method and the hot fermentation process of composting from the point of view of the conservation of manurial constituents and other factors such as hygienic requirements, cost of operation, net income to the municipality etc.

To start with, the writer would like to clear up a certain amount of indefiniteness which seems to persist even at present in the relative use of the terms 'poudrette' and 'compost', in relation to the treatment of nightsoil. Before the principles of compost making became clear as the result of the pioneering work of Hutchinson and Richards [1921], the term poudrette was indiscriminately applied to mixtures of nightsoil with other materials, either organic such as rubbish or inorganic such as soil or ash [Leather, 1895; Kelkar, 1909]. But now the term 'compost' is generally applied to bulky organic materials which have undergone fermentation, usually by the addition of a nitrogenous starter. Since nightsoil is one of the best starters that could be used for this purpose, the term 'compost' would properly apply to the product obtained by fermentation of bulky material such as street sweepings with the addition of nightsoil. The term 'poudrette' is best restricted to the products obtained by dehydration of nightsoil either with dry earth or other materials such as ash, sawdust, lignite or by the action of heat [Bruttini, 1923].

Kinds of poudrettes.—Nightsoil can be converted into poudrette by one of several ways: viz. (1) by simple exposure to the sun in trenches, as in the Nasik system, with a thin covering of rubbish in order to prevent flybreeding; (2) by the addition of woodash in shallow beds and thorough mixing, as in the Poona system [Kelkar, 1909]; (3) by the addition of other drying agents such as powdered lignite, peat, sawdust etc. [Bruttini, 1923] or (4) by the effect of heat, along with the addition of chemicals such as chlorine, sulphuric acid etc. in order to remove the odour [Bruttini 1923]. Of these methods, systems 1 and 2 are simple and are easy of adoption by most municipalities in this country, whereas systems 3 and 4 require either special equipment or require materials in large quantities which are not easily available at most centres. Hence, in the investigations reported in the present paper, particular attention was paid to systems 1 and 2 only mentioned above.

POUDRETTE WITHOUT THE ADDITION OF ANY DEHYDRATING AGENT

Fifteen hundred lb. of nightsoil were led into trenches 2 ft. broad, 3 ft. deep and 5 ft. long. The nightsoil occupied a depth of $2\frac{1}{2}$ ft. in the above trenches and was covered over loosely with a layer of the organic fraction of

street sweepings to a depth of about three inches. Fifty lb. of the organic refuse were used for the purpose. The experiment was carried out in duplicate and observations were made on the changes undergone in the system.

It was noticed that in about 48 hours there was rapid frothing and liquefaction of the mass, as evidenced by the nightsoil separating into a solid layer at the bottom and a thinner opalescent layer at the top. Bubbles of gas were given off from the mass, which burst open the rubbish layer at places; this indicated that probably the lower layers of the mass were undergoing anærobic decomposition. The gases evolved were not analyzed, but tests with litmus and turmeric papers kept in the midst of the rubbish covering the top, showed the evolution of ammonia. This was confirmed by keeping a glass basin containing a known amount of standard dilute sulphuric acid on the rubbish, which absorbed considerable quantities of ammonia.

Portions of the upper liquid layer were taken out, after removing the scum at the top and were analyzed for ammonia on succeeding days, with the results shown in Table II. At the end of a week, the mass rapidly settled down and the upper opalescent liquid layer had slowly disappeared, due probably to seepage into the ground. It is evident that a good portion of the ammonia contained in this layer must have been lost either by diffusion into the air or by seepage into the ground.

TABLE II
Evolution of ammonia from nightsoil in trenches

Days from start	NH ₃ N in 5 c. c. of opalescent upper layer mg.
2	3.6
3	5.2
4	6.8
5	4.6
6	3.2
7	2.1

The mass was allowed to remain in the trench for six months, at the end of which period it had dried completely and got into a powdery condition, partly mixed with the surrounding soil and the rubbish added at the top. The mass still smelt strongly of nightsoil, indicating that the process was more one of drying than of decomposition. The mass was scraped out, weighed and analyzed. The data obtained are shown in Table III. The analytical and sampling procedures were the same as reported in an earlier communication [Acharya, 1939]. In the present case also the resulting manure was contaminated by the addition of a considerable amount (about 94 lb.) of extraneous soil from the trench and outside; a correction has been applied for this soil contamination according to the method reported in an earlier paper [Acharya, 1940] and the corrected figures are shown in column 4 of Table III.

TABLE III
Poudrette from nightsoil

Constituents	Taken at the start 1500 lb. N. S. + 50 lb. rubbish (lb.)	Recovered at the end as poudrette (lb.)	Recovery corrected for soil contamination (lb.)	Percentage recovery of constituents.	Percentage analysis of poudrette (Column 3)
1	2	3	4	5	6
1. Dry matter . . .	337.7	308.8	213.7	63.28	..
2. Ash free organic matter .	256.6	133.8	132.6	51.68	43.33
3. Carbon . . .	154.9	76.8	76.2	49.20	24.87
4. Nitrogen . . .	17.11	7.13	7.08	41.38	2.31
5. Ash . . .	81.1	175.0	81.1	100.00	56.66
6. P_2O_5 . . .	12.18	6.38	6.36	52.23	2.07
7. K_2O . . .	6.86	3.94	3.72	54.23	1.28

The percentage recoveries of the original constituents given in column 5 of Table III, would show that nearly half the manurial constituents and in the case of nitrogen more than half are lost during the process of poudrette making, if the nightsoil be allowed to dry by itself without the addition of soil. Since the losses fall equally on the potash and phosphoric acid as well as on carbon and nitrogen, it is presumed that most of the loss should have been due to seepage into the ground of the liquid fraction formed in the preliminary liquefaction of nightsoil.

The percentage analysis of the final poudrette obtained is shown in column 6 of Table III. The data show that the manure is of good quality inspite of the heavy losses of manurial constituents during the process of poudrette making. The above losses are masked to a certain extent by the small recovery of manure obtained by this method. From 1500 lb. of nightsoil (fresh weight) only about 300 lb. of poudrette are obtained whereas the same quantity of nightsoil would yield about 2000 lb. of dry manure in the form of compost.

As regards the hygienic aspects of the method it was noticed that smell nuisance and flybreeding could not be effectively overcome by the loose cover of rubbish put on top of nightsoil.

NIGHTSOIL-EARTH POUdRETTES

In this method, 1500 lb. lots of nightsoil were let into trenches of the same size as in the preceding experiment, but the nightsoil was covered over

with a layer of dry earth six inches deep, instead of with a thin layer of organic refuse. When cracks appeared on the surface, more soil was added. It was found necessary to add 500 lb. of soil.

The sinking of the poudrette mass was more rapid in this case than in experiment I, probably due to the rapid absorption of water by the soil and its subsequent evaporation from the soil surface. The pressure of the soil layer at the top might have also helped to force down the liquefied portion of the nightsoil into the ground below, quicker. Flybreeding was effectively prevented in this method. The trenches were opened at the end of six months. Undecomposed nightsoil was still present in lumps of black masses and the poudrette smelt strongly of nightsoil. The mass was scooped out of the trench, along with the soil and was weighed and analyzed.

The analytical figures are summarized in Table IV. In addition to the 500 lb. of soil added to cover the nightsoil, further soil to the extent of about 200 lb. had got mixed as extraneous contamination. Due correction has been applied for this and the percentage recoveries of the constituents are given in column 5 of Table IV. These figures are similar to those given in Table III, though the recoveries are slightly better in the present experiment. The higher conservation of nitrogen is probably due to the absorption of ammonia by the soil layer at the top, which otherwise might have been lost into the atmosphere.

TABLE IV
Nightsoil-earth poudrette

Constituents	Taken at the start 1500 lb. N. S. + 500 lb. soil (lb.)	Recovered at the end as poudrette (lb.)	Recovery corrected for soil contami- nation (lb.)	Percent- age recovery of consti- tuents	Percent- age analysis of poudrette on dry basis (Column 3)
1	2	3	4	5	6
1. Dry matter	777.5	846.3	654.1	84.14	..
2. Ash free organic matter .	235.5	114.5	112.1	47.62	13.53
3. Carbon	142.5	.8	58.6	41.14	7.07
4. Nitrogen	16.75	8.71	8.61	51.41	1.03
5. Ash	542.0	741.8	542.0	100.00	87.64
6. P_2O_5	12.0	6.83	6.79	56.62	0.81
7. K_2O	7.5	3.99	3.55	47.43	0.47

The presence of a soil layer at the top improves the hygienic aspects of the method, but does not prevent the loss of the liquid portion of nightsoil by seepage into the ground and the consequent loss of manurial constituents. This can to a certain extent be avoided by interspersing thin layers of nightsoil and dry earth one over the other, but this would mean the addition of much larger quantities of earth than have been added in the present experiment and the consequent increased dilution of the manurial value of the poudrette.

The figures given in column 6 of Table IV would show that even when only 500 lb. of soil are added to 1500 lb. of nightsoil, which corresponds to a six inches layer of soil over a $2\frac{1}{2}$ feet layer of nightsoil, the manurial value of the resulting poudrette is reduced considerably. The product contains barely 1 per cent of nitrogen, 0.81 per cent of P_2O_5 and 0.47 per cent of K_2O and is comparable to an ordinary type of compost; the organic matter content is however lower in the former case. The yield of poudrette in the present instance is about 846 lb. from 1500 lb. of nightsoil (fresh weight). As observed already, about 2000 lb. of dry manure in the form of compost could be obtained from the same quantity of nightsoil. This would show that though the covering over of nightsoil with earth in poudrette-making secures an improvement over using a loose cover of organic refuse, this method cannot compare with the composting process, so far as the full utilization of the manurial constituents of nightsoil is concerned.

NIGHTSOIL-ASH POUURETTE

This system was given an extensive trial by the Poona Municipality in the beginning of this century, before the city was fitted up with underground sewerage. Kelkar [1909], however, remarks that in 1906 the system was given up by the municipal contractor in favour of what at present we would call a method of composting with town sweepings, presumably because the latter method gave a higher yield of manure from the same quantity of nightsoil.

Experiments were first carried out, in the present investigations, on the use of different proportions of nightsoil to wood-ash, and also on the use of coal-ash in place of wood-ash. Large quantities of coal ash are available at some centres, especially near railway workshops and factories. The preliminary experiments showed that when the amount of ash added was less than 40 per cent by weight of nightsoil (fresh) taken, the mass did not become solid. When one part by weight of ash was added to two parts by weight of nightsoil and the whole well mixed, the resulting product was a solid mass which easily dried when exposed to the sun for a day. The above proportion by weight corresponds roughly to a proportion by volume of equal quantities of wood-ash and nightsoil. The poudrette so obtained was ash-grey in colour and possessed, if at all, only a faint trace of odour. Even this trace of smell could be got over by the addition of about 10 per cent of powdered charcoal to the wood-ash.

Coal-ash was found to be equally satisfactory as a dehydrant and deodorizer for nightsoil; but a serious drawback of coal-ash lies in the large amount

of iron and alumina contained in it. It is well known that a high proportion of these is inimical to plant growth and renders unavailable the phosphoric acid present in the manure as well as that already present in the soil.

TABLE V

Mineral composition of nightsoil ash, wood-ash and coal-ash.

Constituents	Nightsoil ash	Wood-ash.	Coal-ash
1. SiO_2 + acid insolubles	23.76	21.45	32.20
2. Fe_2O_3 + Al_2O_3	7.85	12.60	41.82
3. CaO	20.16	33.60	4.59
4. K_2O	10.11	5.30	0.92
5. P_2O_5	18.87	3.18	0.41

The relative composition of the ash constituents present in nightsoil, wood-ash and coal-ash is shown in Table V.

The preliminary experiments were followed by semi-large scale experiments carried out with 1000 lb. lots of nightsoil to which 500 lb. of wood-ash or coal-ash were added. The mixing was done with long handled spades in shallow wide trenches, bricklined and plastered inside. Immediately after mixing, the grey, solid mass was taken out and spread out to dry on brick-lined platforms in the sun for a day or two; after which, the bigger lumps were broken down with a wooden mallet and the manure was ready for being packed in bags for transport. From 100 parts of nightsoil (fresh weight) and 50 parts of ash, about 75 parts by weight of sun dry poudrette, containing about 10 per cent of moisture were obtained.

The chemical composition of the poudrettes obtained by the use of wood-ash and coal-ash respectively is given in Table VI. It will be noted therefrom that the coal-ash poudrette is equal to the wood-ash poudrette in regard to its content of organic matter and of nitrogen, but is much poorer in calcium, potash and phosphoric acid. Field trials with the above two types of poudrettes—to be reported in a subsequent communication—also confirmed the much superior manurial value of the wood-ash poudrette.

TABLE VI

Comparison of the chemical composition of wood-ash and coal-ash poudrettes (analytical figures on dry basis)

Constituents	Poudrette of night-soil with 50 per cent wood-ash	Poudrette of night-soil with 50 per cent coal-ash
	(Per cent)	(Per cent)
1. Ash free organic matter	18.51	18.84
2. Carbon	11.12	11.86
3. Nitrogen	1.32	1.30
4. Ash	81.49	81.16
5. K_2O	4.17	1.11
6. P_2O_5	2.84	1.01
7. CaO	24.24	4.31

The advantages of the ash poudrette method as compared with the earth poudrette method described in section on 'night-soil earth poudrette' above or the natural drying of night-soil examined in section on 'poudrette without the addition of any dehydrating agent' above are: (1) the rapidity with which the process is finished, the ash-poudrette method requiring only two to three days, as against six to eight months needed in the other two cases; (2) the smaller amount of space required, the ash-poudrette method requiring only a shallow tank for mixing operations and a drying yard, whereas the other methods require long trench space; (3) the effective control of smell and flybreeding secured in the ash-poudrette method, so that the poudrette making unit can be situated quite near to towns instead of at a considerable distance from towns as in the other methods; this means considerable saving in cartage expenses; (4) the absence of a loss of manurial constituents in the ash-poudrette method due to volatilization or to seepage into the ground; (5) the considerable increase in the manurial value of the product obtained, due to the addition of wood-ash which is rich in calcium, potash and phosphoric acid; (6) the greater yield of manure obtained by the wood-ash method, as compared to the other two methods. The use of coal-ash in place of wood-ash secures the advantages mentioned above, except No. 5. In fact, the manurial value is adversely affected due to the high concentration of iron and alumina present in coal-ash.

DISCUSSION

In assessing the relative merits of the poudrette vs. composting systems for the disposal of nightsoil, the following points have to be considered from the point of view of the municipality, viz. (a) hygienic considerations of smell

flybreeding and thorough destruction of noxious products ; (b) simplicity of operations and cost of process to the municipality ; and (c) the yield and quality of manure obtained and the net income obtainable by the municipality over and above the cost of processing. It will be convenient to consider these points separately.

(a) *Hygienic considerations*

The Nasik system of covering the nightsoil in trenches with a thin layer of rubbish (*katchra*) would appear to provide scope for flybreeding, unless stringent precautions are adopted to cover up immediately the portions where the nightsoil froths up and bursts the covering, especially in the initial stages. Since the process is mainly one of drying, the nightsoil does not undergo satisfactory decomposition and the resulting poudrette still retains an offensive odour, which becomes perceptible when the material is spread on the land ; the smell is carried in the direction of the wind over several miles even. It is this drawback which militates against the more widespread use of this material in our country—especially in areas where the farmer lives on his land.

The covering of the nightsoil trenches with a thick layer of mud six inches deep or more, satisfactorily prevents flybreeding. The microbial decomposition is greater in this case, due to the sinking down of the soil and its admixture with the nightsoil. But the admixture is not thorough, nor does ordinary earth by itself supply the carbonaceous material necessary for effective decomposition. As such, the decomposition is only partial in this case and the resulting poudrette still smells of nightsoil, though to a lesser extent than in the case where nightsoil is dried by itself. A marked disadvantage of adding earth to nightsoil, for purpose of drying, is the considerable dilution of manurial constituents produced thereby. This point will be dealt with in more detail under manurial considerations.

The ash-poudrette method is a distinct improvement over the other two systems, since wood-ash acts as a satisfactory disinfectant and also removes most of the smell. Flybreeding is successfully prevented. The main drawback of the ash-poudrette method, from the hygienic point of view, is that it does not secure a thorough destruction of the noxious products e.g. through a process of fermentation as the composting process does. The nightsoil ash system is more or less a mechanical mixture—at least for some weeks after its preparation ; and the addition of water is generally enough to separate the constituents and regenerate the undecomposed constituents of nightsoil.

From the above point of view, composting is the most hygienic method of disposal of nightsoil. The material is so thoroughly decomposed in the process, that the resulting product possesses generally a pleasant earthy smell and is devoid of any unpleasant odour. If the hot fermentation system of composting be adopted, flybreeding and smell are most effectively overcome.

(b) *Simplicity of operations and cost*

It is no doubt true that the present system of 'trenching' the nightsoil, as adopted by most municipalities in this country is simple, but it is highly inefficient from the manurial point of view. With a little more control and

supervision, it is possible to compost the nightsoil with street rubbish in the same trenches as are used at present, with the result that a much greater quantity of manure of good quality could be obtained.

The preparation of wood-ash-nightsoil poudrettes, if properly organized, may prove to be the simplest of all methods of disposal of nightsoil. The process requires much less space and time than the 'trenching' method and the cost of operation is proportionately decreased. It may be possible to cut down the time and space required for the ash-poudrette method still further, by carrying out the operations with simple machinery at higher temperatures, e.g. 60-70°C, such that a dry product is obtained straight from the 'mixer', which is ready for immediate packing and transport.

But the ash-poudrette method suffers from one or two serious disadvantages, which detract greatly from the value of its simplicity and cheapness. In the first place, it requires large supplies of wood-ash which may not be readily available. About six to eight tons of wood-ash per day may be required to deal with the nightsoil collected in a town with a population of about 100,000. The Poona Municipality tried to overcome this difficulty [Kelkar, 1909] by burning the organic portion of street refuse and using the ash so obtained. But this method should be considered wasteful, since it involves the loss of organic matter and of nitrogen contained in such street refuse, which possess distinct manurial value on our soils which are poor in organic matter and nitrogen. It would be sounder economics to convert such organic street refuse into manure by a process of fermentation. As an alternative to the method of burning street refuse, for obtaining the ash contained therein, municipalities can have recourse to a systematic collection of house hold ash separately from the dustbins and subjecting this to a preliminary heating before using it for poudrette making. This method may prove practicable in some of the smaller municipalities. At other centres where coal-ash is available in large quantities, this may be used to supplement the available supplies of wood-ash.

(c) Quantity and quality of manure and income to the municipality

Table VII gives data comparing the chemical composition of poudrettes against composts of nightsoil with street sweepings. In assessing the relative merit of any one method of disposal of nightsoil in comparison with another, consideration should be paid not merely to the chemical composition of the manure prepared but also to the total quantity of it obtainable by the method in question. Thus, referring to the data presented in Table VII one would note that the poudrette obtained by simple drying of nightsoil in trenches with a thin covering of rubbish (Nasik system) is very satisfactory in its chemical composition and contains a higher percentage of nitrogen, organic matter and phosphoric acid than composts. But the amount of manure obtained by the method from 1000 lb. of nightsoil is only 270 lb., including the rubbish added and extraneous soil contamination, while the hot fermentation system yields over five times the above quantity of manure (on dry basis). The system adopted by most municipalities of covering the nightsoil in trenches with a heavy layer of soil, yields a larger quantity of manure, viz. 564 lb. than the Nasik system, but in this case the manurial constituents are very much diluted by the inert mass of soil added. In spite of the dilution, the

resulting poudrette contains about 1 per cent of nitrogen and 13.5 per cent of organic matter and should be considered as satisfactory for application to land. But the total quantity of manure obtained by this system is still only a third of what is obtainable by the best systems of composting and the chemical composition is definitely poorer.

TABLE VII

Comparison of the chemical composition and yield of poudrettes and composts

	Poudrette by drying nightsoil with a thin covering of rubbish.	Nightsoil-earth poudrette	Nightsoil-wood-ash poudrette	Compost from N. S. and street rubbish	
				Hot fermentation method	Aerobic method
	lb.	lb.	lb.	lb.	lb.
Yield of manure (dry wt. from 1,000 lb. fresh Night-Soil.	270	564	700	1,500	1,000
Percentage analysis of manure on dry basis	Per cent	Per cent	Per cent	Per cent	Per cent
1. Ash free organic matter .	43.33	13.53	18.51	31.85	28.89
2. Carbon	24.87	7.07	11.12	18.01	14.98
3. Nitrogen	2.31	1.03	1.32	1.13	0.99
4. Ash	56.66	87.64	81.49	68.15	71.11
5. P_2O_5	2.07	0.81	2.84	1.08	1.48
6. K_2O	1.28	0.47	4.17	1.06	1.48
7. CaO	24.24	9.12	12.77

The wood ash-poudrette method on the other hand has the attractive feature that it yields a good type of manure rich in lime, potash, phosphoric acid and nitrogen.

As will be seen from Table VII, the quantities of phosphoric acid, potash and calcium present in the ash-poudrette are two to four times as great as those ordinarily present in composts and the nitrogen percentage is slightly higher. As such it seemed worthwhile making a critical comparison of the economics of disposal of town wastes by the above two methods—on the supposition that the organic portion of street rubbish is incinerated to yield the ash required in the former method. The results of such a comparative

study are presented in Table VIII. Data for the preparation of coal-ash-nightsoil poudrettes are also included, though, as already pointed out, the manurial value of such poudrettes is much less—the reason for the inclusion being that large quantities of coal-ash are available as waste material at several centres.

TABLE VIII

Comparison of the economics of conversion of nightsoil into composts and ash-poudrettes

Data per ton of compost (50 per cent moisture) or per ton of poudrette (10 per cent moisture)	Compost of Night-soil with street sweepings.		Poudrette of Nightsoil with ash	
	Hot fermentation process	Aerobic method	Wood-ash	Coal-ash.
<i>Refuse required.</i>	lb.	lb.	lb.	lb.
Nightsoil	750	1,100	3,000	3,000
Street sweepings	1,250	1,650
Wood or coal-ash	1,500	1,500
	Re. A. P.	Re. A. P.	Re. A. P.	Rs. A. P.
Extra cost to the municipality in preparing compost or poudrette per ton of manure.	0 8 0	1 0 0	0 8 0	0 8 0
	Per cent.	Per cent.	Per cent.	Per cent.
Plant nutrients per ton of manure (50 per cent moisture in composts and 10 per cent in ash poudrettes).	N 0.57 P ₂ O ₅ 0.54 K ₂ O 0.53 CaO 4.56 Organic matter. 15.92	0.49 0.74 0.74 6.38 14.45	1.19 2.83 3.75 21.82 16.66	1.17 1.00 1.18 3.88 16.96
	Rs. A. P.	Rs. A. P.	Rs. A. P.	Rs. A. P.
Price of above nutrients at normal market rates for inorganic fertilizers viz.—				
Rs. 5 per unit of N (1 per cent per ton) . . .	6 12 0	7 8 0	25 8 0	12 8 0
Rs. 25 per unit of K ₂ O and P ₂ O ₅				
Re. 0.1 per unit of CaO.				
Re. 0.05 per unit of organic matter.				
Price of above nutrients calculating on the basis of 50 per cent availability of the nutrients.	3 6 0	3 12 0	12 12 0	6 4 0
Expected sale price per ton of manure	2 0 0	2 0 0	5 0 0	2 8 0
	At Re. 1 per cartload (½ ton) of manure (50 per cent moisture)		Supplied by weight per ton (10 per cent moisture)	
Profit over cost of preparation per ton of manure . . .	1 8 0	1 0 0	4 8 0	2 0 0
	Tons	Tons	Tons	Tons.
Amount of manure that could be prepared in a municipality of 100,000 population.	12,000	8,000	3,000	3,000
	Rs.	Rs.	Rs.	Rs.
Annual income to municipality after deducting extra expenses incurred for manure preparation.	18,000	8,000	13,500	6,000
Approximate annual expenses of municipality of 100,000 population for collection and removal of nightsoil and street rubbish (excluding supervising staff).	36,000	36,000	36,000	36,000

In the data presented in Table VIII only the extra cost to the municipality in carrying out the composting or poudrette making is included. The costs of collection of refuse materials and their transport to the manure making depot are not included, it being assumed that these form part of the regular sanitary work of the municipality which is being carried out at present and will have to be continued in future, purely from considerations of hygiene, irrespective of the way in which the refuse materials are disposed off finally. In comparing the relative costs of processing assigned to poudrette making and to composting, it must be noted the poudrettes prepared contain only about 10 per cent of moisture, whereas the composts contain 40 to 50 per cent of moisture. Hence, on an equal dry-weight basis, the cost of processing in compost making would be almost double the figures given in Table VIII. Between the two systems of composting, the higher charges allotted to the aerobic method are due to : (a) the number of turnings that have to be given in that method and consequent increased labour charges ; (b) the extra water that has to be added in the aerobic method at each turning ; and (c) the larger amount of refuse material that has to be dealt with initially, to produce one ton of final compost, on account of the greater loss of organic matter in the aerobic method.

The content of plant nutrients contained in the different manures (Table VIII) are taken from Table VII, and the price per ton of manure has been calculated on the basis of the normal unit prices for the chief manurial ingredients such as nitrogen, potash, phosphoric acid and calcium contained in the corresponding inorganic fertilizers. The values so obtained represent only the upper limit, for it is well known that the nutrients present in bulky organic manures such as composts or farmyard manure are but partially available for plant growth and that only slowly. The residual value of an organic manure, however, is an important consideration for which due provision must be made in assessing the cost of that manure. Long duration experiments extending over several decades, carried out at Rothamsted and at Woburn [Rothamsted Report, 1932] with farmyard manure have shown that about 50 per cent of the nitrogen of farmyard manure is recovered in the crops and in the soil over a period of years, but the remaining 50 per cent is lost, either into the atmosphere or into the sub-soil. Corresponding field trials with composts in order to assess the nitrogen availability of different types of composts have not been made, but it may be assumed that in the case of composts prepared from nightsoil, the availability of nitrogen would be at least 50 per cent. Actual field trials carried out at Bangalore, over several seasons, have shown that such nightsoil composts and poudrettes give higher crop yields than farmyard manure.

The degree of availability of the other manurial constituents such as potash, phosphoric acid and calcium, present in composts, has not been determined, but taking on a rough average that all constituents are available to an extent of 50 per cent, the prices per ton of manure work out to Rs. 3-6-0 to Rs. 3-12-0 in the case of nightsoil compost, Rs. 12-12-0 for the wood-ash-poudrette and Rs. 6-4-0 for the coal-ash-poudrette.

But, in fixing the prices of the manures, due consideration must be paid to the cost of transport of the manure from the Municipal Depot to the ryot's land, probably several miles off. In order to provide for both the above

factors, the actual sale prices of the manures are fixed in Table VIII at about $\frac{1}{2}$ to $\frac{1}{3}$ of their intrinsic manurial value. Thus, the composts are priced at Rs. 2 per ton, i.e. at Re. 1 per cartload of half a ton, while the wood-ash-poudrette is priced at Rs. 5 per ton and the coal-ash-poudrette at Rs. 2-8-0 per ton.

The profit over the cost of processing, is found to be greatest in the case of the wood-ash-poudrette, being Rs. 4-8-0 per ton, as compared to Rs. 2 per ton obtained for the coal-ash-poudrette, Re. 1-8-0 per ton for the hot fermented compost and Re. 1 per ton for the aerobically prepared compost. But these profits have to be weighed against the respective total quantities of manure that could be prepared in any particular area. In a municipality with a population of 100,000 for instance, about 4,000 tons of nightsoil and 20,000 tons of street sweepings containing about 5,000 tons of organic refuse, may be expected to be collected per year. From this amount of refuse, it would be possible to prepare about 12,000 tons of compost (50 per cent moisture) by the hot fermentation process, 8,000 tons of compost (50 per cent moisture) by the aerobic method and only 3,000 tons of poudrette manure (10 per cent moisture) either with wood-ash or with coal-ash. Multiplying the profit per ton of manure by the respective quantity of manure prepared, we find that the greatest income to the municipality (Rs. 18,000 per year) could be expected by adopting the hot fermentation system of composting, followed respectively by the wood-ash-poudrette method (Rs. 13,000), the aerobic method of composting (Rs. 8,000) and the coal-ash-poudrette method (Rs. 6,000).

It will be interesting to compare the above incomes with the average expenses incurred by a municipality of the above size for the actual collection and transport of nightsoil and street rubbish (excluding the salaries of the supervising staff), which may be estimated at about Rs. 36,000 per year. Hence, by adopting the hot fermentation process of composting, it would be possible for the municipality to recoup about half of the expenses incurred for the collection of town refuse in its area, whereas by the wood ash poudrette method more than a third of the above expenses could be recovered. This income is in addition to what the municipality could obtain by a preliminary sorting out, from street rubbish, of products such as waste paper, rags, tins, leather, iron pieces, glass etc., which could be marketed separately and be made to yield a considerable revenue, especially in the bigger municipalities.

A greater income than the above could be obtained by a judicious combination of both the compost and ash-poudrette methods. It has already been pointed out in an earlier communication [Acharya, 1940] that though the hot fermentation process of composting secures a better conservation of nitrogen than the aerobic method, still about 20-25 per cent of the nitrogen is lost in the former case. This loss was attributed to the narrow C : N ratio of nightsoil (about 8 : 1) and of street sweepings in India (about 25 : 1), and, consequently, of a mixture of the two. It was pointed out also in the above communication that the loss could be minimized by widening the initial C : N ratio, which could be effected to a certain extent by decreasing the proportion of nightsoil : organic refuse : soil fraction below the ratio of 2 : 2 : 1 (by weight) used in the above experiments. This would mean the setting free of a portion of the nightsoil to be disposed off otherwise than by composting. The quantity of nightsoil so liberated could best be converted into wood-ash-poudrette.

It is, therefore, suggested that with a view to secure the fullest utilization of the manurial constituents present in town wastes (including nightsoil, house-hold ash and street sweepings) with the least loss of those constituents during the process of conversion into manure, it would be of advantage to a municipality to adopt and carry on both systems of manure-making side by side, viz. preparation of ash-poudrette and composting with street sweepings. The quantity of nightsoil used for poudrette making would depend on the quantity of ash, (household ash or coal-ash) available in the locality; and the remaining bulk of nightsoil could be composted with street sweepings, preferably in a proportion of not less than one part by weight of nightsoil for every two parts of the 'organic' fraction and one part of the 'soil' fraction of street sweepings.

An objection sometimes raised against the adoption of the composting procedure is that it requires too much of space and time. But this difficulty will not arise in those cases (forming over 90 per cent of the municipalities in India) where the 'trenching' (or 'pitting') system is at present being adopted for the disposal of nightsoil, since the operations of composting could be carried on in a fraction of the trench area used at present and in a much shorter time. But in the case of densely populated urban areas of big cities, provision of enough space for composting purposes may offer practical difficulties. In such cases, it is possible to minimize considerably the space and time required for composting by carrying on the process in closed cells at a rapid rate in (about 15 days) with the help of suitable mechanical devices, as in the Hygienic Process recently adopted by the Kensington Borough Council [Anstead, 1939] for dealing with its town refuse. But the relative economics of such mechanized processes, as compared to the simpler types of composting or poudrette making described in this paper, require further and more detailed examination, especially under Indian conditions of labour and market prices for organic manures, before the adoption of such mechanized systems could be recommended.

SUMMARY

1. A critical study has been made of the different methods of poudrette preparation, e.g. by use of (a) nightsoil without the addition of earth; (b) nightsoil *plus* earth and (c) nightsoil *plus* ash, in comparison with the methods of composting, with special reference to: (a) hygienic considerations; (b) manurial value of the products obtained, as revealed by chemical composition; (c) cost of operation and (d) total income obtainable by a municipality by adopting the process.

2. It is found that the drying of nightsoil by itself involves heavy losses of nitrogen, organic matter, phosphoric acid and potash, due to liquefaction of nightsoil and seepage of the liquid portion into the ground. The addition of dry earth decreases the losses to some extent, but as considerable quantities of earth are required, the resulting product of nightsoil with earth is low in manurial constituents.

3. The preparation of wood-ash-nightsoil poudrettes has several features to commend it, from the hygienic and manurial standpoints, and is a promising method of disposal of nightsoil. The operations can be carried out in a compact plant, quite rapidly, saving space and time and yielding a product

of high manurial value, which could be readily transported and sold over a wider area than compost.

4. The preparation of wood-ash-nightsoil poudrettes, however, suffers from some serious drawbacks, viz. (a) the difficulty of obtaining enough supplies of ash, and (b) the fact that the method does not solve the problem of a satisfactory disposal of street rubbish. The total yield of manure obtained by composting street rubbish with nightsoil is nearly twice as much, on the dry basis, as is obtainable by the conversion of nightsoil into the ash poudrette; and the net income to the municipality by adopting the composting process is nearly $1\frac{1}{2}$ times as much as in the other case.

5. With a view to secure the advantages of both systems, it is recommended that the system of poudrette making from nightsoil and wood-ash may be adopted to the limit of the local availability of wood (or house-hold) ash and that the remaining bulk of nightsoil may be composted with street rubbish.

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NOTES

NOTICE 1 OF 1940 (JANUARY, MARCH 1940)

THE following plant quarantine regulations and import restrictions have been received in the Imperial Council of Agricultural Research. Those interested are advised to apply to the Secretary, Imperial Council of Agricultural Research, New Delhi, for loan.

LIST OF U. S. DEPARTMENT OF AGRICULTURE, BUREAU OF ENTOMOLOGY AND PLANT QUARANTINE, SERVICE AND REGULATORY ANNOUNCEMENTS.

1. *Quarantine and other official announcements* :—
Dutch Elm Disease Quarantine Regulations—modifications.
2. *Summaries of plant quarantine import restrictions* :—
 - (i) *Kingdom of Italy—Italian East Africa*—cotton restrictions.
 - (ii) *Republic of Paraguay—Basic Legislation*—the digest.
 - (iii) *Republic of Turkey*—Prohibited plant pests and diseases.
 - (iv) *Colony and Protectorate of Kenya*—Government notice No. 468—addition of potatoes to the list of restricted seeds.
 - (v) *Jamaica. British West Indies*—Import permit required for plant material.
3. *Service and Regulatory Announcements*—
 - (i) *April—June 1939.*
 - (ii) *July—September 1939.*
4. *List of Intercepted Plant Pests—1938.*

CHANGES IN NOMENCLATURE

WITH the transfer of the Imperial Agricultural Research Institute from Pusa to New Delhi it has been found necessary to alter the nomenclature of the 'Pusa' varieties. The improved varieties so far evolved at Pusa and others that may in future be bred at New Delhi will henceforth be known as 'Imperial Pusa' varieties. The number of the variety in each case will be preceded by the letters 'I. P.'. This nomenclature will also be adopted for the milch herd of the Institute as well as for herbarium specimens and specimens of insects, fungi, etc. This change has been made to keep up the earlier association of the Institute with the word 'Pusa' and at the same time to distinguish the strains bred by the Imperial Department of Agriculture from those which may be bred by the Bihar Agricultural Department at their station at Pusa.

A list of the old and the new names of the varieties of crops under distribution is given below :—

Crop	Old name	New name
Wheat	Pusa	4
Do.	"	12
Do.	"	52
Do.	"	80—5
Do.	"	111
Do.	"	114
Do.	"	120
Do.	"	125
Do.	"	165
Barley	Type	13
Do.	"	21
Oats	B. S.	1
Do.	B. S.	2
Do.	Hyb.	C
Do.	"	G
Do.	"	J
Paddy	Pusa Type	9
Do.	"	18
Do.	"	24
Do.	"	31
Do.	"	52
Do.	"	124
Do.	"	129
Do.	"	144
Rahar.	"	15
Do.	"	24
Do.	"	51
Do.	"	64
Do.	"	80
Gram.	"	2
Do.	"	6
Do.	"	17
Do.	"	25
Do.	"	28
Do.	"	53
Do.	"	58
Mung	"	18
Do.	"	23
Do.	"	28
Do.	"	36
Urid	"	4
Do.	"	6
Do.	"	7
Do.	"	14
Lentil	"	11
Do.	Hyb.	111-86
Peas	S.	29
Linseed	Type	12
Do.	"	121
Do.	"	124
Linseed	Pusa H.	10
Do.	H.	21
Do.	H.	55
Do.	H.	68

Crop	Old name		New name	
Sesamum . . .	Pusa Type . . .	3	I. P. . . .	3
Do.	" "	7	I. P.	7
Do.	" "	29	I. P.	29
Safflower . . .	" "	30	I. P.	30
Chilli	" "	34	I. P.	34
Do.	" "	41	I. P.	41
Do.	" "	46—A	I. P.	46—A
Do.	" "	51	I. P.	51
Hemp	" "	3	I. P.	3
Do.	" "	6	I. P.	6
Do.	New Hibiscus .		I. P. Sab. . .	5
Tobacco (<i>N. Tabacum</i>) . . .	Pusa Type . . .	28	I. P.	28
Do.	" "	58	I. P.	58
Do.	" "	63	I. P.	63
Do.	" Hyb. . . .	142	I. P. Hyb. . .	142
Tobacco (<i>N. rustica</i>) . . .	" Type	18	I. P.	18
Indian Hemp . .	" "	1	I. P.	1

REVIEW

**Supplement to Root Nodule Bacteria and Leguminous Plants, BY E. B. FRED.
I. L. BALDWIN & E. MCCOY**

THE authors have collected the references to the recent work and have listed the papers published from 1932, the date of publication of their monograph 'Root Nodule Bacteria and Leguminous Plants', upto 1938. A few papers which were overlooked in the authors' book in 1932 are also listed separately. The index is also supplemented by a list of scientific names of all plants cited in the original monograph and by an author index. No attempt is made in this supplement to interpret the results of the recent investigations. The supplement will be useful as a comprehensive list of references to workers in this field of research. [N. V. J.]

AA

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2. All communications regarding subscription and advertisements should be addressed to the Manager of Publications, Civil Lines, Delhi.

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